Canine leishmaniasis in an endemic region, Northeastern Brazil: a comparative study with four groups of animals

Matheus Resende Oliveira1 · Manuel Benicio Oliveira Neto1 · Taynar Lima Bezerra2 · Weslania Souza Inacio da Silva1 · Wandklebson Silva da Paz3 · Igo Gonçalves dos Santos1 · Márcio Bezerra-Santos3,4,5 · Victor Fernando Santana Lima1,3

Received: 26 July 2021 / Accepted: 13 September 2021 / Published online: 9 October 2021
© The Author(s), under exclusive licence to Springer-Verlag GmbH Germany, part of Springer Nature 2021

Abstract
Canine leishmaniasis (CanL) is a zoonosis caused by protozoa of the genus *Leishmania* and remains an important public health concern in tropical areas. In Brazil, domestic dogs are considered the most relevant reservoir of the parasite and one of the main targets of the disease control actions. Considering this, we aimed herein to evaluate the CanL infection in different canine groups and distribution of cases in the state of Sergipe, an endemic region in Northeastern Brazil. The evaluated 467 animals were classified into four groups: hunting (*n* = 50), company (*n* = 64), guard (*n* = 140), and wandering (*n* = 213). Samples (blood, bone marrow, conjunctival swab, and lymph node aspirate) were collected from animals in nine municipalities of Sergipe. First, all animals were submitted to general and ophthalmic clinical examination. Next, they were tested serologically by TR-DPP®, and for the presence of *Leishmania*, amastigotes in samples of bone marrow, conjunctival swab, and lymph node aspirate were diagnosed by PCR and parasitological techniques. It was observed that 34.69% (162) of the evaluated dogs were seropositive. The highest rates of positivity were found in hunting 54% (27/50; OR = 3.52; *p*-value = 0.001) and guard dogs 42.14% (59/140; OR = 2.18; *p*-value = 0.01). Otherwise, the highest percentage of symptomatic dogs was observed in wandering animals (85%; OR = 9.63; *p*-value < 0.0001). The distribution of case analysis showed that the highest positivity rates occurred in inland municipalities situated in arid regions. Taken together, our data demonstrate that hunting and guard dogs are among the animals most exposed and affected by clinical manifestations of CanL, mainly in the inland municipalities of Sergipe State.

Keywords *Leishmania* · Wandering dogs · Diagnosis · Epidemiology

Introduction
Leishmaniasis is considered one of the most important public health concerns in the world, especially in tropical and poor areas of Africa, Asia, and Latin America (WHO 2020). They comprise a set of parasitic diseases, of a zoonotic character and broad clinical spectrum, responsible for annually affecting approximately 700,000 to 1 million people worldwide (Torres-Guerrero et al. 2017; Sundar and Singh 2018). Clinically, they are classified as tegumentary (TL) or visceral leishmaniasis (VL), and this last one can be lethal in 90% of cases if left untreated (Sousa et al. 2015a, b; Menezes et al. 2016). Despite the involvement of wild animals (foxes, opossums, and rodents) in the transmission of *Leishmania*, domestic animals, such as the canine species, are considered the main reservoir of the parasite and source of infection in...
Canine leishmaniasis is caused by intracellular protozoa of the genus *Leishmania*. In Brazil, the disease has been related to eight species of parasites, but especially *L. (Leishmania) infantum* and *L. (Viannia) braziliensis* (Rocha et al. 2015; Ferreira et al. 2018). The infection occurs mostly through the bite of female insects of the genus *Lutzomyia* (Diptera: Psychodidae), including *L. longipalpis* as the main vector in the country (Santos et al. 2019; Galvis-Ovallos et al. 2020). In addition, although considered a rural endemic, the process of urbanization of leishmaniasis has been observed in Brazilian capitals and municipalities, such as the city of Aracaju-SE (Campos et al. 2017; Reis et al. 2017; Silva et al. 2017a, b, c). Factors such as adaptive capacity of sandflies, cities expanding to forest areas, and the presence of the domestic dog are mostly related to these changes in the epidemiological profile of the disease (Nunes et al. 2016; Reguera et al. 2016).

When infected, dogs usually express high cutaneous parasitism, which facilitates ingestion by sandflies and maintenance of the parasite’s transmission circle (Otranto and Dantas-Torres 2013). Clinically, they may present in the most advanced stages onychoglyphosis, lymphadenomegaly, apathy, weight loss, eye damage, alopecia, and dermatitis. Otherwise, many animals can be asymptomatic, with an infection rate that can reach 80% in some places (Eguchi et al. 2017; Contreras et al. 2019). Additionally, as they are the main urban reservoirs for leishmaniasis and being some risk of transmission for humans, dogs have become the main targets of disease control actions in Brazil (Shang et al. 2011; Nunes et al. 2016).

Currently, different techniques for canine leishmaniasis (CanL) diagnosis are recommended by the Brazilian Ministry of Health (Ministério da Saúde 2016). The main screening method for CanL is the DPP Immunochromatographic Test (Dual-Path Platform, Biomanguinhos®). The immunoenzymatic assay-ELISA (EIE Canine Visceral Leishmaniasis, Biomanguinhos®) is used as a confirmatory test for seroreagent animals. Additionally, these tests can be associated with complementary tools for parasitological diagnosis, according to the stage of the disease, such as the search for amastigotes in biological samples and the molecular biology tests by polymerase chain reaction (PCR) (Ministério da Saúde 2016; Silva et al. 2016).

Notably, Brazil is the country responsible for the leishmaniasis endemic disease in the Americas (about 96% of the cases occur here) and it has historically been neglected. Regardless, surveys that assess the situation of CanL are still scarce and punctual in a few municipalities (Cordeiro et al. 2016; Santos and Sousa 2018). In addition, due to the wide territorial distribution, leishmaniasis is still worrying in Northeastern Brazil, which reinforces the need for studies that comprehend the current panorama of the disease and help in development of effective control strategies (Ribeiro et al. 2019; Lamoumier et al. 2020). More importantly, Sergipe is one of the states with the highest disease positivity rate and still with related deaths. Additionally, a study conducted by Campos and colleagues Campos et al (2017) demonstrated an increasing time trend of cases of canine and human leishmaniasis in the capital Aracaju, between 2008 and 2014. Considering this, we aimed to evaluate CanL infection in four canine groups and the spatial distribution of cases in the state of Sergipe, an endemic region in Northeastern Brazil.

### Material and methods

#### Study type and ethical aspects

An ecological time series study was conducted, using spatial analysis techniques, and assessing the seroprevalence of CanL in four different groups of dogs. The spatial analysis units consisted of municipalities from the state of Sergipe, Northeastern Brazil. The Ethics Committee on Animal Use (CEUA), from the Universidade Federal de Sergipe, approved all procedures herein performed (License number: 1003190520).

#### Study area

The present study was conducted in rural and urban areas of nine municipalities, located in the metropolitan/coast and inland regions of Sergipe (Supplementary Material 1). The state is divided into climatic zones semiarid and subhumid (Caatinga and Atlantic Forest), with annual rainfall index ranging from 500 to 1000 mm (SEAGRI-SE, 2020). Importantly, most of the population lives in the urban area (73.52%). Regardless, Sergipe has precarious socioeconomic indicators in Brazil: the average monthly household income is R$ 1028.00 per capita; the Human Development Index (IDH) is 0.665 (below the national average), and there is lack in the basic sanitation services (IBGE, 2020). Annually, there are about 70 cases of human visceral leishmaniasis in the state of Sergipe (SINAN, 2020).

#### Animals and sampling

From November to December 2020, a total of 467 dogs from different groups (company (*n* = 64); guard (*n* = 140); hunting (*n* = 50); and wandering (*n* = 213)), of both genders and different ages, were sampled. The animals were selected, randomly and by non-probabilistic convenience, obtaining the largest number of sample/dog. The sample characterization is shown in Table 1. Initially, animals were physically
evaluated, and all data recorded in clinical charts. Afterwards, biological samples were obtained. We collected blood samples from all assessed animals \((n = 467)\). Additionally, conjunctival swab from both eyes of dogs with ophthalmic manifestations was collected. Fine needle aspiration of lymph nodes in those animals presenting enlargement of this organ was performed. Finally, bone marrow aspiration was performed in dogs with at least three clinical alterations compatible with CL and reagents for anti-Leishmania antibodies in immunochromatographic test Dual-Path Platform TR-DPP® (Biomanguinhos®, Rio de Janeiro, RJ, BR).

### Laboratory analysis

All blood samples obtained were stored in test tube containing ethylenediaminetetraacetic acid (EDTA) and maintained at 6 ºC. We proceeded with the serological analysis using immunochromatographic test Dual-Path Platform TR-DPP® (Biomanguinhos®, Rio de Janeiro, RJ, BR). This test detects IgG anti-Leishmania antibodies through recombinant antigens K28 and Protein A conjugated to colloidal gold and adsorbed into nitrocellulose membrane.

Next, microscopic slides were prepared using the material obtained from the conjunctival swab, bone marrow aspirate, or lymph node. Each slide was stained using the rapid method panoptic (Laborclin), and they were analyzed by two professionals using optical microscopy \((40×\text{and} 100×)\).

Lastly, DNA was extracted from bone marrow samples using a commercial kit (Kit Go Taq® Green Master Mix, Promega, Madison, WI, USA) according to the manufacturer’s instructions. The amplification of fragment of 447 bp of the \(L.\ infantum\) was performed using the primers MC1: 5‘-GTTAGCCGATGTTGCTTCTTGTCTTG-3‘ and MC2: 5‘-CAC CCATTGTTTCCGATTCTTG-3‘ (Cortes et al. 2004). The amplifications were analyzed after 1% agarose gel electrophoresis in UV transilluminator.

### Statistical analysis

Data were statistically analyzed for absolute and relative frequencies. The chi-square test with Yates’s correction was used to compare differences between groups. The significance level was set at 5% and the differences between the groups were considered statistically significant when a \(p\)-value \(<0.05\) was obtained. All analyses were performed using the software GraphPad Prism version 8.0.1.

### Spatial analysis

To map the distribution of positive dogs, we first obtained the digital cartographic mesh (in shapefile format) of Sergipe State, segmented by municipalities, in the Geographic Projection System latitude/longitude. We obtained this mesh from the databases of the Brazilian Institute of Geography and Statistics (IBGE, 2020).

Posteriorly, maps representing positive cases of CanL were constructed. For that, we used the positivity rate considering the group of dogs: company, hunter, guard, wandering, and all of them together (total). Thereafter, the results were represented on thematic maps stratified according to the following parameters of positivity for CanL: low (0.02 to 7.22%); medium (7.23 to 16.29%); high (16.3 to 29.31%); intense (29.32 to 51.88%); and very intense (51.89 to 100%) (PAHO/WHO, 2017). Herein, we used the QGIS software, version 3.4.11 (QGIS Development Team; Open Source Geospatial Foundation Project) for generating the choropleth maps.

### Results

Study in the Northeast area observed that 34.69% \((n = 162/467)\) of the positive dogs were positive in the screening test for anti-Leishmania antibodies (TR-DPP®) (Table 2). Amastigotes of Leishmania spp. were observed in 29.55% of bone marrow samples, in 25% of the of lymph node aspirates, and in 6.55% of the conjunctival swab of samples obtained from dogs reactive to CanL. In addition, \(L.\ infantum\) DNA was detected in 22.73% of dog samples submitted to PCR. Interestingly, a higher percentage of positives animal in males (47.54% vs. 26.41%; OR = 2.52; \(p\)-value < 0.0001) was observed, but no difference was seen comparing animals from urban and rural areas (34.68% and 34.69%, respectively; \(p\)-value > 0.99).

Thereafter, to compare the positivity for CanL considering the categories of dogs evaluated in the study, the chi-square statistical test was applied and observed a significant difference between them \((p\)-value = 0.0004).
Among the positive animals in one or more diagnostic tests, we identified symptoms for CanL in 56.18% of them. Furthermore, the presentation of symptoms between the groups and observed statistical difference was compared (*p*-value < 0.0001). Considering that the hunting group had the lowest percentage of symptomatic animals (37.04%), we used it as a comparison parameter (Table 4). Statistical differences when comparing with the guard (38.98%; OR = 1.08; *p*-value = 0.99) and company groups were not found (43.75%; OR = 1.32; *p*-value = 0.75). On the other hand, a significant difference when compared to the wandering group was observed (85%; OR = 9.63; *p*-value < 0.0001).

Concerning the main clinical manifestations in positive animals, alopecia (29.01%), onychogryphosis (22.22%), exfoliative dermatitis (20.37%), lymphadenomegaly (14.81%), and ear tip lesions (12.26%) were observed. Additionally, the identified eye disorders were compatible with canine eye leishmaniasis in 9.88% (*n* = 16) of dogs. Mainly, periocular alopecia (6.25%), blepharitis (12.5%), conjunctivitis (81.25%), keratitis (37.5%), eye discharge (68.75%), and corneal ulcer (6, 25%) were observed.

Analysis of case distribution data showed a very intense positivity rate of CanL in inland municipalities of the state. In addition, municipalities in the metropolitan area, including the capital Aracaju, had an intense positivity rate (Fig. 1). Maps in Fig. 2A–D show the spatial distribution of the positivity rate of CanL according to the groups of dogs evaluated. Inland municipalities with very intense positivity rate for CanL in the group of hunting dogs were identified (Fig. 2B). Likewise, guard and wandering dogs showed very intense and intense rates for CanL in inland municipalities and in the metropolitan area, respectively (Fig. 2C, D).

**Discussion**

In this study, the groups of dogs most exposed to *Leishmania* infection in endemic areas of the state of Sergipe, Northeastern Brazil, were identified. The positivity of animals observed here was higher than those obtained in

---

### Table 2

| Laboratory analysis technique                        | Positive *n* (%) | Negative *n* (%) | Odds ratio | 95% CI   | *p*-value |
|------------------------------------------------------|------------------|------------------|------------|----------|-----------|
| Serology (TR-DPP®)                                   | 162 (34.69)      | 305 (65.31)      |            |          |           |
| Amastigotes in bone module                           | 13 (29.55)       | 31 (70.45)       |            |          |           |
| Amastigotes in lymph node                            | 11 (25)          | 33 (75)          |            |          |           |
| Amastigotes in optical conjunctiva                   | 3 (6.25)         | 45 (93.75)       |            |          |           |
| PCR (DNA for *L. infantum*)                          | 10 (22.73)       | 34 (77.27)       |            |          |           |

*n*, number of sample; *PCR*, polymerase chain reaction; DNA, deoxyribonucleic acid

---

### Table 3

| Group of dogs | Positive *n* (%) | Negative *n* (%) | Odds ratio | 95% CI   | *p*-value |
|---------------|------------------|------------------|------------|----------|-----------|
| Company*      | 16 (25)          | 48 (75)          | 1.00       |          |           |
| Wandering     | 60 (28.17)       | 153              | 1.17       | 0.62 to 0.74 | 0.74      |
| Guard         | 59 (42.14)       | 81 (57.86)       | 2.18       | 1.15 to 4.13 | 0.01      |
| Hunting       | 27 (54)          | 23 (46)          | 3.52       | 1.6 to 7.65 | 0.001     |

*n*, number of samples

*Considering that the company group had the lowest percentage of positivity, it was used as a base group for the statistical analysis by chi-square test

Herewith, the group that presented the lowest percentage of positivity (company = 25%) was used as a parameter for comparison with the others (Table 3). Interestingly, hunting dogs were the ones that had the highest odds ratio for CanL (54%; OR = 3.52; *p*-value = 0.001), along with guard dogs (42.14%; OR = 2.18; *p*-value = 0.01). Surprisingly, no differences were observed between company and wandering groups (28.17%; OR = 1.17; *p*-value = 0.74).

---

### Table 4

| Group of dogs (*n* CanL +) | Clinical classification | Odds ratio | 95% CI   | *p*-value |
|----------------------------|-------------------------|------------|----------|-----------|
|                           | Symptomatic *n* (%)     | Asymptomatic *n* (%) |          |           |
| Hunting* (*n* = 27)        | 10 (37.04)              | 17 (62.96) | 1.00     |           |
| Guard (*n* = 59)           | 23 (38.98)              | 36 (61.02) | 1.08     | 0.4 to 2.71 | 0.99     |
| Company (*n* = 16)         | 7 (43.75)               | 9 (56.25)  | 1.32     | 0.36 to 4.62 | 0.75     |
| Wandering (*n* = 60)       | 51 (85)                 | 9 (15)     | 9.63     | 3.31 to 25.29 | <0.0001 |

*n*, number of samples; CL +, positive for canine leishmaniasis; CI, confidence interval

*Considering that the hunting group had the lowest percentage of animals presenting signs and symptoms, it was used as a base group for the statistical analysis by chi-square test
epidemiological surveys carried out in other states, such as Bahia (Rodrigues et al. 2020), Mato Grosso (Carvalho et al. 2020), Minas Gerais (Castro-Júnior et al. 2014), and São Paulo (Julião et al. 2007). These results may be related to the diversity of biological samples and laboratory tests used in our study, which may allow an increase in the rates of sensitivity for the LC diagnosis (Sassaki et al. 2011; Braz et al. 2016).

Conversely, these findings corroborate a worrying and insidious scenario in relation to leishmaniasis in Sergipe. Considering that canine infection usually precedes human cases, the high number of positive dogs identified here, distributed in different regions and municipalities of the state, represents a high risk of parasite transmission to human patients and maintenance of the epidemiological circle of the disease (Campos et al. 2017).

Currently, the Brazilian Ministry of Health recommends, in epidemiological surveys, the use of Dual-Path Platform canine visceral leishmaniasis (DPP® CVL)—rapid test that uses a combination of recombinant antigens rK39, K26, and K9—for the detection of anti-Leishmania antibodies in animals (Grimaldi Júnior et al. 2012; Ministério da Saúde 2016). Regardless of the recommendation of more sensitive tests, such as ELISA and RIFI as confirmatory tests, it is worth noting that the sensitivity of DPP® CVL can be greater than 98%. This test is recommended for screening dogs infected with CanL, and in our study, it is demonstrated that its use, along with parasitological examination and PCR, can be highly useful for Leishmania sp. diagnosis in surveys (Grimaldi Júnior et al. 2012; Paltrinieri et al. 2016; Lins et al. 2018; Sales et al. 2020).

Several studies in the literature aim to assess the influence of biological factors, such as sex and age, on the positivity rate for leishmaniasis (Silva et al. 2016; Campos et al. 2017; Evaristo et al. 2020). Herein, it was observed that male dogs were mostly affected by the disease in Sergipe (OR = 2.52). According to Silva et al. (2016) and Evaristo et al. (2020), the prevalence of male dogs infected in the semi-arid region of the Northeast is about 2.25 times higher than that in females, mainly in animals of 12 and 84 months of age.

Similarly, a study conducted by Campos and colleagues Campos et al (2017) in Aracaju identified a higher percentage of CanL among males. Remarkably, in the epidemiology of CanL in the Northeastern Brazil, the levels of rainfall, the occurrence of natural ecotones, and the presence of forest in the peri-urban regions are factors associated with Leishmania transmission and, as a result, both sexes are equally exposed (Almeida et al. 2012; Santos and Sousa 2018). Taken together, these findings demonstrate that, despite studies, the relationship between sex and susceptibility to CanL is still not fully understood, and further research is required, especially on the influence of biological factors.

Concerning the groups of dogs most affected by CanL, Almeida and colleagues Almeida et al (2012) point out that
animals living outside (hunting, guard, and wandering dogs) are more accessible to sandflies and therefore more exposed to infection by *Leishmania* sp. Corroborating this, our data showed that hunting and guard dogs were mostly affected by CanL and had higher odds ratio of infection by the parasite (OR = 3.52 and 2.18, respectively), when compared to company dogs. This may be related to the environment in which these animals are kept, since many of these dogs live adjacent to the feeding and breeding sites of *Lutzomyia* females (chicken coops, pig pens, corrals, and barns). Moreover, these animals are exposed to forest areas during hunting activities, usually in twilight and post-twilight periods, coinciding with the highest activity of sandflies (Carvalho et al. 2010; Dantas-Torres et al. 2010; Gálvez et al. 2010; Piantedosi et al. 2016; Aklilu et al. 2017; Campino and Maia 2018; Silva et al. 2018a, b; Trájer et al. 2018; Lago et al. 2020).

Notably, several studies report that wandering dogs are the most affected by CanL. Nonetheless, a statistical difference here when comparing the positivity rate between wandering and company groups was not observed. On the other hand, considering the seropositive animals, wandering dogs were those mostly diagnosed with signs and symptoms (85%) and odds ratio of 9.63. This significantly higher
odds ratio in this group may be related, first, to the habit of these animals roaming the streets, peripheral, and forest areas (places of exposure to sandflies). Additionally, this may be linked to nutritional factors, such as protein-calorie deficiency, which is much more common in this group. As a result, it directly affects the general health of these animals, compromises immunity, and facilitates the development of the disease, and the clinical presentation of signs and symptoms (Melo et al. 2018; Lewgoy et al. 2020; Nweze et al. 2020).

Concerning the clinical aspects, among symptomatic dogs, classic clinical alterations of leishmaniasis such as alopecia, onychogryphosis, lymphadenomegaly, and ear tip lesions were observed (Moreira et al. 2016; Contreras et al. 2019; Azevedo and Marcili 2020; Pasanisi 2020). Furthermore, the ocular form of CanL was also observed in these dogs, in which changes such as periorcular alopecia, blepharitis, keratitis, and conjunctivitis are among the most reported ophthalmic and clinical manifestations are consequences of systemic impairment caused by Leishmania, which involves direct mechanisms of tissue damage due to the presence of the parasite and specific cytotoxic immune responses. Clinical manifestations are consequences of systemic impairment caused by Leishmania, which involves direct mechanisms of tissue damage due to the presence of the parasite and specific cytotoxic immune responses (Peña et al. 2008; Brito et al. 2010; Rálic and Jovanovic 2011; Pietro et al. 2016).

Importantly, our study had some limitations that deserve to be mentioned. First, the sample number was lower among guard and hunting animals. Nevertheless, this is expected, as this population of dogs is usually smaller. Second, some difficulties in accessing communities in rural areas for data collection and sampling were faced. Finally, collections due to the COVID-19 pandemic and the need for social distance had to be the limitations. For this reason, other municipalities in the state were not contemplated. In spite of this, an expressive sample number was obtained (n = 426) which, divided among the study groups and along with the statistical and spatial analysis, allowed to identify the categories most exposed to the infection and to map the spatial dynamics of the leishmaniasis in the state.

Conclusion

Altogether, our results showed that hunting and guard dogs are among the animals most exposed and affected by clinical manifestations of CanL, mainly in the inland municipalities of Sergipe State. This corroborates the occupational risks involved in the transmission of the parasite. Additionally, although street animals are the main targets of seroepidemiological surveys for CanL, our data clearly point to the need to routinely assess other canine groups, such as guard and hunting dogs, especially in endemic areas. Importantly, it was observed that, among positive animals, the wandering group is the one with the most clinical signs and symptoms for leishmaniasis. Thereby, this requires national public policies for the epidemiological monitoring and care of these animals. Furthermore, this study aimed to improve the diagnosis of CanL, reduce the risk of *Leishmania* transmission, and control the disease in the country.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s00436-021-07319-0.

References

Aklilu E, Gebresilassie A, Yared S, Kindu M, Tekie H, Balkew M, Warburg A, Hailu A, Gebre-Michael A (2017) Comparative study on the nocturnal activity of phlebotomine sand flies in a highland and lowland foci of visceral leishmaniasis in north-western Ethiopia with special reference to *Phlebotomus orientalis*. Parasit Vectors 10:1–10

Almeida ABPF, Sousa VRF, Cruz FACS, Dahroug MAA, Figueiredo FB, Madeira MF (2012) Canine visceral leishmaniasis: seroprevalence and risk factors in Cuiabá, Mato Grosso, Brazil. Rev Bras Parasitol Vet 21:359–365

Azevedo RCF, Marcili A (2020) Alterações cutâneas secundárias à infecção por *Leishmania* sp.: revisão de literatura. Braz J Develop Sci 6:19328–19346

Braz PH, Lima MAM, Maia JS (2016) Sensibilidade parasitológica de diferentes locais de colheita para o diagnóstico de leishmaniose em cães. Arq Ciênc Vet Zool 19:91–93

Brito PLC, Laus JL, Tafuri WL, Figueiredo MM, Silva Júnior VA, Maia FCL, Alves LC (2010) Histopathological findings and detection of parasites in the eyes of dogs infected naturally with *Leishmania chagasi*. Cienc Rural 40:1141–1147

Campino L, Maia C (2018) The role of reservoirs: canine leishmaniasis. In: Ponte-Sucre A, Padrón-Nieves M (Eds), Drug resistance in *Leishmania* parasites: consequences, molecular mechanisms and possible treatments. Cham, Switzerland, 380 pp.

Campos R, Santos M, Tunon G, Cunha L, Magalhães L, Moraes J, Ramalho D, Lima S, Pacheco JA, Lipscomb M, Jesus AR, Almeida RP (2017) Epidemiological aspects and spatial distribution of human and canine visceral leishmaniasis in an endemic area in northeastern Brazil. Geospat Health 12:67–73

Carvalho MR, Dias AFR, Almeida ABF, Alves MR, Paes AS (2020) Canine visceral leishmaniasis: perception, prevalence, and spatial distribution in municipality of Nossa Senhora do Livramento, Mato Grosso, Brazil. Rev Bras Parasitol Vet 29:1–11

Carvalho SMS, Santos PRB, Lanza H, Brandão-Filho SP (2010) Diversity of *Phlebotomus* sp. in the municipality of Ilhéus, Bahia. Epidemiol Serv Saúde 19:239–244

Castro-Júnior JG, Freire ML, Campos SPS, Scopel KKG, Porrozzi R, Silva ED, Colombo FA, Silveira RCV, Marques MJ, Coimbra ES (2014) Evidence of *Leishmania* (*Leishmania*) *Infantum* infection in dogs from Juiz de Fora, Minas Gerais state, Brazil, based on immunochromatographic Dual-Path Platform (DPP®) and PCR assays. Rev Inst Med Trop São Paulo 56:225–229

Contreras IK, Machado MA, Rocha COJM, Oliveira GR, Carvalho FCG (2019) Sinais clínicos apresentados por cães positivos para leishmaniose visceral no município de Vassouras, Rio de Janeiro. PUBVET 13:1–6
Cordeiro LLLR, Silva EM, Lima VFS, Porto WJN (2016) Leishmaniose visceral canina na microrregião serrana dos quilombos, leste alagoano, Alagoas, Brasil. Hygeia Rev Bras Geo Med Saúde 12:169–176

Cortes S, Rolão N, Ramada J, Campino L (2004) PCR as a rapid and sensitive tool in the diagnosis of human and canine leishmaniasis using Leishmania donovani s.l.-specific kinetoplastid primers. Transactions of the Royal Society of Tropical Medicine and Hygiene 98(1):12–17. https://doi.org/10.1016/S0035-9203(03)00002-6

Dantas-Torres F, Andrade AJ, Tenório KER, Andrade Filho JD, Bal-Lewgoy B, Mastrangelo A, Beck L (2020) Tanatopolítica e biossegurança: dois regimes de governo da vida para a leishmaniose visceral canina no Brasil. Horiz Antropol 26:145–176

Lins TNB, Souza IB, Barros GMMR, Santos CVB, Silva RP, Alves LC, Carvalho GA, Ramos RAN (2018) Seroprevalence and spatial distribution of canine leishmaniasis in an endemic region in Brazil: how has the situation changed after 10 years? Rev Soc Bras Med Trop 51:680–682

Melo SN, Teixeira-Neto RG, Werneck GL, Struchiner CJ, Ribeiro RAN, Sousa LR, Melo MOG, Carvalho Júnior CG, Penaforte KM, Manhani MN, Aquino VV, Silva ES, Belo VS (2018) Prevalence of visceral leishmaniasis in a population of free-roaming dogs as determined by multiple sampling efforts: a longitudinal study analyzing the effectiveness of euthanasia. Prev Vet Med 161:19–24

Menezes JA, Luz TCB, Sousa FF, Verne RN, Lima FP, Margonari C (2016) Fatos de risco peridomiciliares e conhecimento sobre leishmaniose visceral da população de Formiga, Minas Gerais. Rev Bras Epidemiol 19:362–374

Ministério da Saúde (2016) Manual de Vigilância, Prevenção e Controle de Zoonoses. Normas técnicas e operacionais. Brasília, Distrito Federal, 123 pp

Moreira NB, Almeida ADBPF, Pinto AZL, Mutzemberg ER, Godoy I, Silveira MM, Dutra V, Sousa VRF (2016) Canine visceral leishmaniasis: dermatological aspects and associated dermatoses. Acta Sci Vet 44:1–4

Navea-Pérez HM, Díaz-Sáez V, Corpas-López V, Merino-Espinosa G, Morillas-Márquez F, Martín-Sánchez J (2015) Leishmania infantum in wild rodents: reservoirs or just irrelevant incidental hosts? Parasitol Res 114:2363–2370

Nunes JB, Laurenzi MD, Kanamura HY, Pereira AAC, Colombo FA, Marques MJ (2016) Leishmania infantum infection in dogs from the southern region of Minas Gerais state, Brazil. Rev Inst Med Trop São Paulo 58:1–7

Nweze JA, Nweze EI, Onoja US (2020) Nutrition, malnutrition, and leishmaniasis. Nutrition 73:1–14

Otranto D, Dantas-Torres F (2013) The prevention of canine leishmaniasis and its impact on public health. Trends Parasitol 29:339–345

Pan American Health Organization (2017) Leishmaniasis: epidemiological report in the Americas. Leish Report 5:1–8

Pasanisi CC (2020) Inquérito epidemiológico sobre a leishmaniose visceral canina no Municipio de Itapevi: relato do primeiro inquérito sorológico. PVBET 14:1–7

Peña MT, Naranjo C, Kluss G, Fondevila D, Leiva M, Roura X, Davidson MG, Dubielzig RR (2008) Histopathological features of ocular leishmaniosis in the dog. J Comp Pathol 138:32–39

Piantedosi D, Veneziano V, Di Muccio T, Manzillo VF, Fiorentino E, Scalone A, Neola B, Di Prisco F, D’ Alessio N, Gradoni L, Oliva G, Gramiccia M (2016) Epidemiological survey on Leishmania infection in red foxes (Vulpes vulpes) and hunting dogs sharing the same rural area in Southern Italy. Acta Parasitol 61:769–775

Pietro S, Di Bosco VRF, Crinó C, Francaviglia F, Giudice E (2016) Prevalence, type, and prognosis of ocular lesions in shelter and owned-client dogs naturally infected by Leishmania infantum. Vet World 9:633–637

Rálic M, Jovanovic M (2011) Ocular manifestation in canine leishmaniosis. Vet Glasnik 65:269–276

Reguera RM, Morán M, Pérez-Péterto Y, García-Estrada C, Balañafouca R (2016) Current status on prevention and treatment of canine leishmaniasis. Vet Parasitol 227:98–114

Reis LL, Balieiro AAS, Fonseca FR, Gonçalves MFJ (2017) Changes in the epidemiology of visceral leishmaniasis in Brazil from 2001 to 2014. Rev Soc Bras Med Trop 50:638–645

Ribeiro CR, Gonçalves CA, Cruz LM, Galera PD (2019) Prevalence of visceral canine leishmaniasis and co-infections in perurban region in the Federal District - Brazil. Cienc Anim Bras 20:1–8

Rocha TJM, Barbosa ACA, Santana EPC, Calheiros CML (2015) Aspectos epidemiológicos dos casos humanos confirmados de...
leishmaniose tegumentar americana no Estado de Alagoas, Brasil. Rev Pan-Amaz Saúde 6:49–54

Rodrigues TF, Benitez AN, Sevá AP, Okamura LH, Galvão AB, Gomes JF, Bresciani KDS, Cardoso TC (2020) Spatial and seroepidemiology of canine visceral leishmaniasis in an endemic Southeast Brazilian area. Rev Soc Bras Med Trop 53:1–7

Sales KGDS, Miranda DEO, Paiva MHS, Figueredo LA, Otranto D, Dantas-Torres F (2020) Fast multiplex real-time PCR assay for simultaneous detection of dog and human blood and Leishmania parasites in sand flies. Parasit Vectors 13:1–10

Santos EFN, Sousa IF (2018) Análise estatística multivariada da precipitação do estado de Sergipe através dos fatores e agrupamentos. Rev Bras Climatologia 23:1–18

Santos JML, Dantas-Torres F, Mattos MRF, Lino FRL, Andrade LSS, Souza RCA, Brito FLC, Brito MEF, Brandão-Filho SP, Simões-Mattos L (2010) Prevalence of anti-Leishmania spp antibodies in dogs from Garanhuns, in the middle scrub zone (Agreste) of Pernambuco. Rev Soc Bras Med Trop 43:41–45

Santos WS, Ortega FD, Alves VR, Garcez LM (2019) Flebotomíneos (Psychodidae: Phlebotominae) de área endêmica para leishmaniose cutânea e visceral no nordeste do estado do Pará, Brasil. Rev Pan-Amaz Saúde 10:1–18

Sassaki CY, Colodel MM, Ferreira I, Nogueira FS, Lucheihs SB, Langoni H, Rocha NS (2011) Comparison of different diagnostic tests in dogs uninfected and naturally infected with visceral leishmaniasis. J Venom Anim Toxins Incl Trop Dis 17:348–352

Secretaria de Estado da Agricultura, Desenvolvimento Agrário e da Pesca de Sergipe, 2020. Clima - Caracterização do Estado. https://www.seagri.se.gov.br/informacoes/1/caracterizacao-do-estado [Accessed 20 Sep 2020]

Shang L, Peng W, Jin H, Xu D, Zhong N, Wang W, Wu Y, Liu Q (2011) The prevalence of canine Leishmania infantum infection in Sichuan Province, southwestern China detected by real time PCR. Parasit Vectors 4:1–5

Silva GI, Silva ET, Costa GRT, Santos IB (2018a) Vigilância da Leishmaniose Visceral no Distrito Federal: aspectos organizacionais, situação epidemiológica e medidas intersetoriais. Comun Em Ciências Da Saúde 28:149–157

Silva JD, Melo DHM, Costa JAG, Costa DF, Silva RBS, Melo MA, Azevedo SS, Alves CJ (2017a) Leishmaniose visceral em cães de assentamentos rurais. Pesq Vet Bras 37:1292–1298

Silva RBS, Mendes RS, Santana VL, Souza HC, Ramos CPS, Souza AP, Andrade PP, Melo MA (2016) Aspectos epidemiológicos da leishmaniose visceral canina na zona rural do semi-árido paraibano e análise de técnicas de diagnóstico. Pesq Vet Bras 36:625–629

Sousa NS, Linhares CB, Pires FGB, Teixeira TC, Lima JS, Nascimento MLO (2015a) Perfil epidemiológico dos casos de leishmaniose visceral em Sobral-CE de 2011 a 2015. SANARE 17:51–57

Sundar S, Singh OP (2018) Molecular diagnosis of visceral leishmaniasis. Mol Diagn Ther 22:443–457

Trájer A, Tânczos B, Hammer T, Padisák J (2018) Solar radiation and temperature conditions as the determinants of occurrence of Phlebotomus neglectus Tonnoir (Diptera: Psychodidae). J Entomol Res Soc 20:13–27

World Health Organization (2020). Leishmaniasis. https://www.who.int/news-room/fact-sheets/detail/leishmaniasis [Accessed 02 Oct 2020]

Publisher’s note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.