ISSN: 0975-833X

Available online at http://www.journalcra.com

International Journal of Current Research
Vol. 12, Issue, 02, pp.10087-10091, February, 2020

DOI: https://doi.org/10.24941/ijcr.37908.02.2020

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

RESEARCH ARTICLE

SEROPREVALENCE OF CANINE LEISHMANIASIS IN PARTS OF SOKOTO STATE, NORTHWESTERN NIGERIA

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ARTICLE INFO

Article History:
Received 14th November, 2019
Received in revised form
20th December, 2019
Accepted 09th January, 2020
Published online 28th February, 2020

Key Words:
Canine, Leishmaniasis,
ELISA, Seroprevalence,
Sokoto.

ABSTRACT

Dogs have been identified as the main reservoirs of *Leishmania* in many parts of the world and play a very important role in the maintenance of the parasite and its transmission to humans and other susceptible hosts. This study was conducted with objective of determining the seroprevalence of canine leishmaniasis in three selected Local Government Areas (Wamakko, Sokoto South and Kware) of Sokoto state, Nigeria. Blood samples were collected from 316 dogs and the separated sera were tested for *Leishmania* spp IgG antibodies using Indirect Enzyme Linked Immunosorbent Assay (ELISA). Total number of seropositive dogs was 11 showing overall seroprevalence of 3.5 % all from the adult dogs. Based on sex, female seropositive dogs account for 3.8% as against males (3.0%). Hunting dogs recorded higher seroprevalence (3.9%) compared to companion dogs (1.7%). There was no significant association between age, sex, use of dogs and seroprevalence of canine leishmaniasis ($P > 0.05$). Highest seroprevalence was recorded in Wamakko L.G.A (5.9%), followed by Sokoto South L.G.A (2.7%) and the lowest was recorded in Kware L.G.A (2.6%). There was statistically significant association ($P < 0.05$) between seroprevanlence of canine leishmaniasis and dog location in the three L.G.A. From this study, we have been able to show evidence of the presence of canine leishmaniasis in parts of Sokoto State, Nigeria. This poses serious possibility of increased risk of the disease transmission to both human and animal population in the study area.

Citation: ***Usman, M., Natala, A.J., Jatau, I.D., Ogo, N.I., Balogun, E.O., Alayande M.O. and Mahmuda, A.*.. 2020.** "Seroprevalence of Canine Leishmaniasis in Parts of Sokoto State, Northwestern Nigeria". *International Journal of Current Research*, 12, (02), 10087-10091.

INTRODUCTION

Leishmaniasis is a disease caused by flagellated protozoan parasite of the genus *Leishmania. Leishmania* species are obligate unicellular parasites that exist in two distinct forms. They exist as non-flagellated amastigotes in humans and other hosts while in culture and gut of sand flies (vector), the flagellated or the promastigote form is visible. Domestic and wild dogs, small rodents are the most important animal reservoir hosts of leishmaniasis, which is transmitted among canines and to humans causing three forms of the disease (visceral, cutaneous, and mucosal) according to the localization of the parasites in mammalian tissues (Travi *et al*., 2018). Of the three forms, visceral leishmaniasis is the most important, with clinical signs such as; alopecia, anemia, epistaxis, hyper-gammaglobulinaemia, generalized lymphadenopathy, skin lesions ulceration, and desquamation of skin and generalized nodular lesions or pustules, chronic renal failure and eventual death (Calvaliero *et al*., 1999). However, dogs that recover after treatment or those that are asymptomatic continue to serve as reservoirs of infection for the phlebotomine sand flies which easily adapt to the peridomestic environment or human dwellings and feed frequently (Killick-Kendrick, 1999; Feliciangeli, 2004). Some known species of *Leishmania* include *Leishmania tropica complex (L. major, L.tropica, L. aethiopica), Leishmania donovani complex (L.d. donovani, L.d. infantum, L.d. chagasi), Leishmania mexicana complex (L.m. amazonensis, L.m. garnhami, L.m. pifanoi) Leishmania braziliensis complex (L.b. braziliensis, L.b. guyanensis, L.b.* panamensis) (Arfan and Simeen, 2008). It has been proven that infected dogs are sources of infection for phlebotomine sand flies, the main vectors of the aetiological agent of zoonotic visceral leishmaniasis (Hommel, 1999; Desjeux, 2004). These phlebotomine sand flies can easily adapt to the peridomestic environment or human dwellings and feed frequently on dogs (Killick-Kendrick, 1999; Feliciangeli, 2004). The common serological methods for the detection of Leishmaniasis are

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Indirect Fluorescent Antibody Test (IFAT), Enzyme Linked Immunosorbent Assay (ELISA) and Direct Agglutination Test (DAT) (Rajasekariah *et al*., 2001; Mikaeili *et al*., 2007). ELISAs based on soluble promastigote or amastigote antigens seem most suited for the serological diagnosis of canine of *Leishmania* infections in both symptomatic and asymptomatic dogs. IFAT though a gold standard, lacks sensitivity in asymptomatic cases but are highly specific. Factors associated with the seroprevalence of leishmaniasis in dogs such as individual traits of dogs (height, sterilization, long fur and age class) have been found to positively affect prevalence while other factors (weight, body score, presence of ectoparasites) and environmental and management features (free ranging or confined dogs) are not associated with seropositivity (de Almeida *et al*., 2014). In Nigeria the endemicity of Canine Leishmaniasis is not known. However, the only available publication on the disease reported a seroprevalence rate of 14.63% in Kwara state, Nigeria (Adediran *et al*., 2016). It is interesting to note that Human Cutaneous Leishmaniasis have been reported in Sokoto state (Jiya *et al*., 2007; Faleke *et al*., 2008) and other parts of Nigeria (Okwori *et al.,* 2001., Salami *et al*., 2004., Adediran *et al*., 2014), yet no information exist on the presence of Canine leishmaniasis in dogs in Sokoto state and its environs. This study therefore was aimed at determining the seroprevalence, distribution and public health impact of Canine Leishmaniasis using Indirect Enzyme Linked Immunosorbent Assay in parts of Sokoto state, North Western Nigeria.

MATERIALS AND METHODS

Study Area: The study area was three Local Government Areas (Wamakko, Sokoto South and Kware) within the Northern senatorial zone of Sokoto State. Sokoto State is located in the semi- arid region of North Western Nigeria between latitude 13° 5′N and longitude 5° 15′E (Mahmuda *et al*., 2012). It shares borders with Niger Republic to the North, Kebbi State to the South West and Zamfara State to the South East. The State has a total land mass of 32,000km^2. The state is characterized by two distinct seasons, the short rainy season which runs from May or June to September or 9 October, depending on the pattern of rainfall of the year. The long dry season starts from October till May or June (Anon, 2001). The minimum relative humidity is less than 20% for most part of the year and ambient temperature ranges from 22^0 C to 43^{0}C (Iloeje, 1971). This was a cross sectional study that covered 3 LGA within the Northern senatorial district of Sokoto state, Nigeria (Figure 1). Blood samples were collected from a total of 316 companion and hunting dogs within the study area. Demographic factors considered during sampling were age (young <1 year, adult ≥ 1 year), sex (male and female), use/purpose of dog (hunting or companion) and location (rural or urban).

Ethical statement: Ethical approval was given by the Sokoto State Ministry of Animal Health and Fisheries Development and sampling was carried out with the consent of the dog owners.

Blood sample collection and serological analysis: Blood samples (4ml) were collectedvia the cephalic vein of each dog into plain bottles without anticoagulant, between May and November 2016, covering both rainy and dry seasons. It was transported to the laboratory on ice and then centrifuged at 1500 rpm for 10 minutes.

The separated sera were transferred into microfuge tubes and stored at -20^{0}C until analysed. The sera were analysed using indirect Enzyme Linked Immunosorbent Assay (ELISA) technique to detect the antibodies (IgG) against Leishmaniasis. A commercial ELISA kit (ID Screen Canine Leishmaniasis, ID-Vet Company, France) was used and the tests were carried out according to manufacturer's instructions based on the principles of absorbance. Samples were read at 450nm using microplate ELISA reader and the mean values (OD) were recorded. The proportion rate of each sample over positive control was calculated by the formula below:

$$SP = \frac{OD\ (sample) - OD\ (NC)}{OD\ (PC) - OD\ (NC)} \times 100$$

SP= Sample to positive ratio
OD = Mean value
PC = Positive control
NC = Negative control

Samples were interpreted as positive if the rate was greater than or equal to 50 %. The ratio greater than 40 % and less than 50 % was considered doubtful and less or equal to 40 % was seen as negative.

Statistical analyses: Data obtained were analysed using SPSS version 20. Chi square (χ^2) and Fisher's exact tests were conducted to test for association between seroprevalence of canine leishmaniasis and age, sex, use, and location of dogs. Values of $P < 0.05$ were considered significant.

RESULTS

Out of 316 studied dogs, only 11(3.5%) were positive for *Leishmania* antibody. Out of the 37 young dogs sampled, none was found to be seropositive while 11 out of the 279 adult dogs were seropositive (Table 1). There was no association between seropositivity and age (**P** >0.05). Only one adult companion dog out of the 11 was seropositive (Table 2) but the frequency of sampled dogs was not equal in different age groups and this could be a confounding factor (Table 1). Highest prevalence was recorded in hunting dogs (3.9%) than companion dogs (1.7%). There was no association (P >0.05) between seroprevalence and use/purpose of dogs (Table 2). A total of 182 (57.6%) female and 134 (42.4%) male dogs were sampled (Table 3). Out of 182 female dogs sampled 7 (3.8%) were positive while 4 male dogs were positive out of 134 (3.0%) sampled. There was no statistical association in seroprevalence between sexes (P > 0.05) (Table 3).Dogs from 3 selected LGA's of Sokoto state were sampled thus: 85 (26.9%) from Wamakko, 75 (23.7%) from Sokoto south and 156 (49.4%) from Kware L.G.A (Table 4). Dogs from WamakkoLGA were most affected with a seroprevalence of 5.9 %, followed by Sokoto South L.G.A with seroprevalence of 2.7 % and Kware LGA recorded a 2.6 % seroprevalence (Table 4). There was significant association (P < 0.05) between seroprevalence and location of sampled dogs (Table 4).

DISCUSSION

Leishmaniasis is transmitted through the bites of sand flies which are more prevalent in warm climatic and arid areas such as Sokoto state. The presence of other predisposing factors such as indiscriminate waste disposal, abundance of termitaria, rodent burrows which has been identified as breeding site of the phlebotomine sand flies (Mascari *et al*., 2013) may be responsible for the results obtained in dogs in the study area.

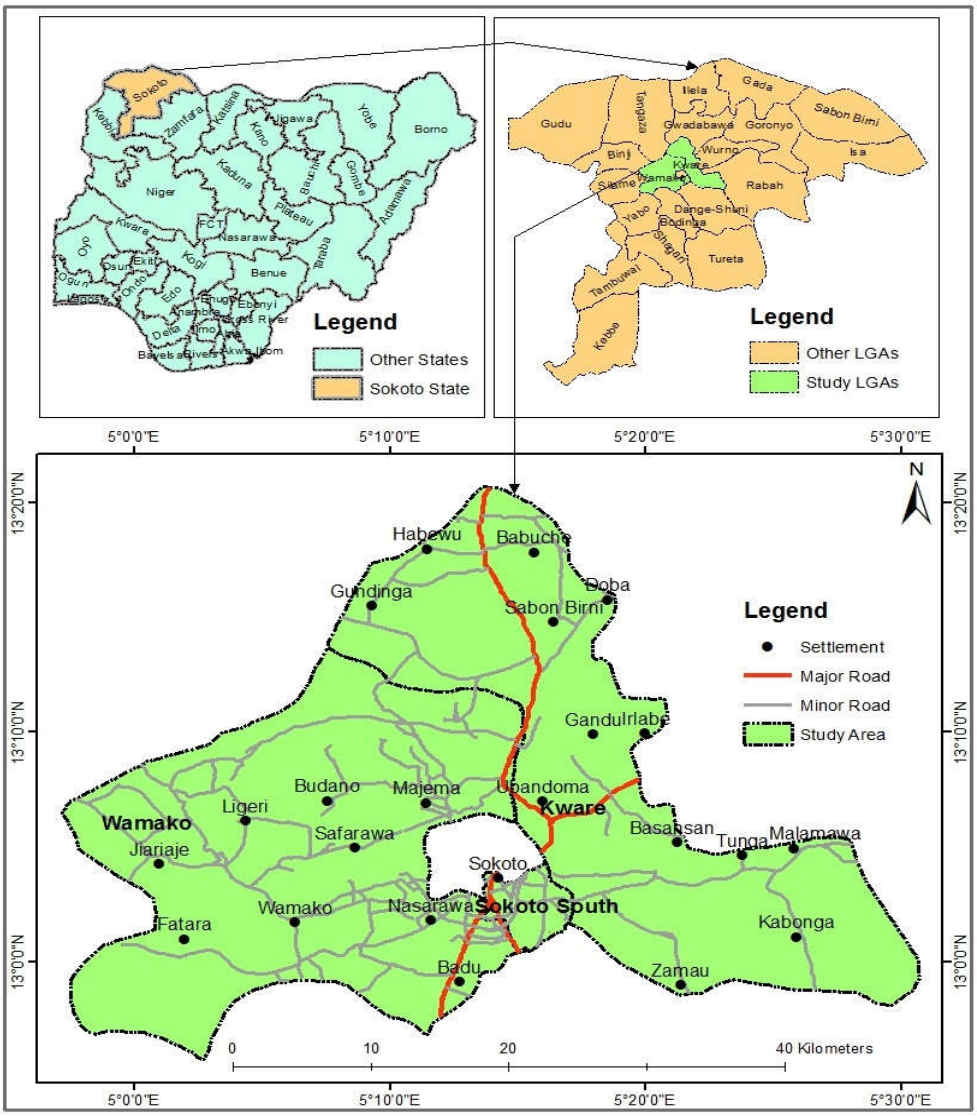

Figure 1. Map of Sokoto state showing the study area

Table 1: Seroprevalence of Canine Leishmaniasis in parts of Sokoto state by Age Group. (N=316)

Age group	Positive n (%)	Negative n (%)	Total n (%)
Adult	11 (3.5)	268 (84.8)	279 (88.3)
Young	0 (0.0)	37 (11.7)	37 (11.7)
Total	11 (3.5)	305 (96.5)	316 (100.0)

FET= 0.000, P> 0.05

Table 2. Seroprevalence of Canine Leishmaniasis in Hunting and Companion Dogs in parts of Sokoto State (N=316)

Use	Positive n (%)	Negative n (%)	Total n (%)
Hunting	10 (3.9)	246 (96.1)	256 (81.0)
Companion	1 (1.7)	59 (98.3)	60 (19.0)
Total	11(3.5)	305 (96.5)	316 (100)

$\chi^2 = 0.726$, P > 0.05

Table 4. Seroprevalence of Canine Leishmaniasis in parts of Sokoto State According to Location (N=316)

Location	Positive n (%)	Negative n (%)	Total n (%)
Wamakko	5 (5.9)	80 (94.1)	85 (100.0)
Kware	4 (2.6)	152 (97.4)	156 (100.0)
Sokoto South	2 (2.7)	73 (97.3)	75 (100.0)

$\chi^2 = 13.807$, P < 0.05

ELISA is one of the serological methods recommended for screening and surveillance studies for canine leishmaniasis, others include IFAT and direct agglutination test (DAT) (Mikaeili *et al*., 2007). In this study, ELISA technique detected the presence of antibodies (IgG), which is by far the most predominant leishmania-specific antibody as observed by Solano *et al.* (2001). High antibody titres correlate with clinical leishmaniasis or indicate a possible manifestation of canine leishmaniasis in infected asymptomatic dogs. Moreover, the specificity and sensitivity of the ELISA test which was used for the detection of Canine Leishmaniasis in this study was reported to be 99.1 and 98.5 % respectively. In a study conducted by Solano *et al*. (2014), ID screen ELISA had a superior diagnostic performance than other serological kits e.g Lei scan, thus, ID screen may be considered as a valuable serological screening test for Leishmaniasis. Seropositivity was higher in hunting dogs than companion dogs which might be due to the fact that hunting dogs are more exposed to sand fly bites as a result of their constant activity in the wild. Kumthekar *et al*. (2014) reported free roaming of dogs as a factor for acquiring L*eishmania* infection. This is similar to the findings of Adediran *et al.* (2014) and it indicates that hunting dogs could play an important role in the epidemiology and transmission of this parasite in the area.

Companion dogs in this study area were not strictly confined, thus can have access to the sand fly breeding sites and eventually their bites. This could explain the reason for the observed seropositivity in the companion dogs which could as well play an important role in the transmission cycle of the disease in the study area. In this study, female dogs showed higher seroprevalence rate compared to the males and may be attributed to the fact that more female dogs were sampled as against males due to the preponderance of female dogs in the study area. Findings from this study suggested that seropositivity for canine leishmaniasis was not associated with age despite all seropositive dogs being in the adult age group.

This could be due to the fact that only 37 of the 316 dogs sampled were young (≤ 1 year old). However, adult dogs are more likely to be used for hunting and other purposes which increase the risk of their exposure to the bites of the sand fly vector. Among the three sampled L.G.As, Wamakko had the highest seroprevalence (5.9%) and this could be due to abundance of refuse dumps in the area. Refuse dumps are good alternative breeding sites for sand flies (Mascari *et al*., 2013) and therefore, their abundance could favour the occurrence of sand flies in an area hence, increasing the rate of *Leishmania* transmission. Similar observation was documented in a previous study on human cutaneous leishmaniasis in Sokoto state (Jiya *et al*., 2007; Faleke *et al*., 2008). The lowest seroprevalence of 2.6% was recorded in Kware L.G.A inspite of having the highest number of dogs sampled and this could be due to the obvious scarcity of sand fly breeding sites (refuse dumps and sewage tanks) in the area.

Conclusion

The overall seroprevalence of canine leishmaniasis in Sokoto state was 3.5% (11/316).Higher seroprevalence was recorded in hunting dogs (3.9%) than companion dogs (1.7%) which indicates that hunting dogs play a greater role in the epidemiology of this parasite as against companion dogs. In addition to determination of *Leishmania* infection, seroprevalence in the domestic dog population could be a helpful way to follow the progress of the disease in endemic areas. There is also need for wider surveillance of the disease in dogs and other suspected reservoir hosts such as rodents in order to ascertain their role in the maintenance of the parasite.

Conflicts of interest: The authors declare that they have no competing interests

REFERENCES

Adediran, O.A., Kolapo, T.U & Uwalaka, E.C., 2014. Seroprevalence of canine Leishmaniasis in Kwara, Oyo and Ogun states of Nigeria. *Journal of Parasitic Diseases,* **1:**1-5.

Anon, 2001. Sokoto State - the seat of the Caliphate, In: Sokoto State Government diary. Pp 2-8.

Arfan, U.I & Simeen, B.R., 2008. Cutaneous Leishmaniasis: An overview of parasitology and host-parasite-vector inter relationship. *Journal of Pakistan Association of Dermatologists*, **18** (42-48).

Cabral, M., O'Grady, J.E., Gomes, S., Sousa, J.C., Thompson, H & Alexander, J., 1998. The immunology of canine leishmaniasis: strong evidence for a developing disease spectrum from asymptomatic dogs. Veterinary Parasitology,**76**:173–180.

Chang, K., Fong, D & Bray, R., 1985. Biology of *Leishmania* and leishmaniasis. In: Chang kp, bray rs, eds. Leishmaniasis: *Human parasitic diseases*. Philadelphia: Elsevier, **1**: 1-30.

Cavaliero, T., Arnold, P., Mathis, A., Glaus, T., Hofmann-Lehmann, R., Deplazes, P., 1999. Clinical,serologic and parasitologic follow-up after long-term allopurinol therapy of dogs naturally infected with *Leishmania infantum*. *Journal of Veterinary Internal Medicine*,**13**(4): 330-334.

De Almeida Curi, N.H., Paschoal, AMd., Mascara, R.L., Marcelino, A.P., Ribeiro, A.A., Passmani, M., Dometrio, G.R & Chiarello, A.G., 2014. Factors associated with the seroprevalence of Leishmaniasis in dogs living around Atlantic forest fragments. *PLoS ONE,***9**(8):e104003.

De Oliveira, L.C.P., de Araujo, R.R., Alves, C.R., Mouta-confort, E., Lopez, J.A & de Mendonca-Lima, F., 2010. Seroprevalence and risk factors for canine visceral Leishmaniasis in the endemic area of Dias D'Avila, state of Bahia, Brazil. *Revista da Sociedade Brasileira de Medicina Tropical*, 43(4)

Desjeux, P., 2004. Leishmaniasis: current situation and new perspectives. *Comparative Immunology, Microbiology and Infectious Disease*, 27:305–318.

Faleke, O.O., Lawal, M.D & Magaji, A.A (2008). Cutaneous Leishmaniosis: Some aspects of epidemiology and a case report. *Sokoto Journal of Veterinary Sciences,* 7(1): 13-16.

Feliciangeli, M.D., 2004. Natural breeding places of phlebotomine sand flies. *Medical and Veterinary Entomology,* 18:71-80.

Hommel, M.,1999. Visceral Leishmaniasis: Biology of Parasite. *Journal of Infection,* 39: 101-11.

Iloeje, N.P. 1971. Where is Nigeria and who are Nigerians? In: A New Geography of Nigeria, Longman Nigeria Ltd, Nigeria. Vol. I., Pp14

Jiya, N.M., Ahmed, H., Jibrin, B & Philips, A.O., 2004. An outbreak of cutaneous Leishmaniosis in a boarding school in Sokoto: Clinical presentation and outcome. Abstracts of Proceedings. *Nigerian Journal of Paediatrics*, 31(3): 104.

Killick-Kendrick, R., 1999. The biology and control of phlebotomine sandflies. *Clinical Dermatology*, 17: 279–289.

Kumthekar, S., Chikweto, A., Chawla, P., Tiwari, K.P., Gozlan, J., Quentin, L., Lanza, M., Paterson, T & Sharma, R.N.,2014. Seroprevalence of Canine Leishmaniasis in Owned and Stray Dogs from Grenada, West Indies. *Journal of Animal Research*. 4(1):131-139

Mascari, T.M., Hanafi, H.A., Jackson, R.E., Ouahabi, S &Ameur, B., 2013. Ecological and control techniques for sand flies (Diptera: *Psychodidae*) associated with rodent reservoirs of leishmaniasis. *PLoS Neglected Tropical Diseases,* 7:2434.

Mahmuda, A., Mohammed, A. A., Alayande, M. O., Magaji, A. A., Salihu, M. D., Fabiyi, J.P., Lawal, M.D., Yakubu, Y., Usman, M. and Danmaigoro, A. 2012. Seasonal prevalence of gastrointestinal nematodes of calves in Sokoto, North-western Nigeria. *ScientificJournal of Veterinary Advances*, 1 (3): 82-89

Mikaeili, F., Hatam, G.R., Fakhar, M., Sarkari, B &Motazedian, M.H., 2007. Comparison of serological methods (ELISA, DAT and IFAT) for diagnosis of visceral leishmaniasis utilizing an endemic strain. Iranian Journal of Immunology, 4:116–121.

Okwori, E.N.A., Nock, I.H., Galadima, M & Ibrahim, S., 2001. Prevalence of cutaneous leishmaniasis in parts of Kaduna State, Nigeria. *Journal of Protozoology Research,* 11: 32-46.

Pourquier, P., Lesceu, S., Dereure, J & Keck, N., 2007. Validation of a new commercial ELISA kit for the detection of directed against Leishmania infantum in canine sera. *World Association of Veterinary Laboratory Diagnosticians–13th International Symposium, Melbourne, Australia*

Rajasekariah, G.H., Ryan, J., Hillier, R., Scott, R & Lisa, P.,2001. Optimisation of an ELISA for the serodiagnosis of visceral leishmaniasis using in vitro derived promastigote antigens. Journal of Immunological Methods, 252:105–119.

Salami, A.K., Oluboyo, P.O., Onile, B.A & Odedina, E.A.,2004. Kala azar in a Nigerian: Report of a case with fatal outcome. *African Journal of Clinical and Experimental Microbiology*, 5(1): 40-45.

Solano-gallego, L., Piera, C., Roura, X., Iniesta, L., Gallego, M & Valladares, J.E., 2001. *Leishmania infantum*-specific IgG, IgG1, and IgG2 antibody responses in healthy and ill dogs from endemic areas. Evolution in course of infection and after treatment. *Veterinary Parasitology*,9(4):256-276.

Solano-Gallego, L., Villanueva-Saz, S., Carbonell, M., Trotta, M., Furlanello, T& Natale, A., 2014. Serological diagnosis of canine leishmaniosis: comparison of three commercial ELISA tests (Leiscan®, ID Screen® and *Leishmania* 96®), a rapid test (Speed Leish K®) and an in-house IFAT. *Parasites & Vectors*,7:111.

Travi, B.L., Cordeiro-da-Silva, A., Dantas-Torres, F., Miró, G., 2018. Canine visceral leishmaniasis:diagnosis and management of the reservoir living among us. *PLoS Neglected Tropical Diseases*, 12(1): e0006082.

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