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Original Article

First serological investigation of *Toxoplasma gondii, Neospora caninum, Leishmania infantum* and *Leptospira* spp. in dogs from a Fulni-ô Indigenous community in Pernambuco, Brazil: a One Health perspective

Primeira investigação sorológica de *Toxoplasma gondii*, *Neospora caninum*, *Leishmania infantum* e *Leptospira* spp. em cães de uma comunidade indígena Fulni-ô em Pernambuco, Brasil: uma perspectiva de Saúde Única

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Abstract

Zoonotic diseases pose a major public health threat in indigenous communities due to geographic isolation, inadequate sanitation, and close contact between humans, domestic animals, and wildlife. This study aimed to investigate the presence of antibodies against *Toxoplasma gondii*, *Neospora caninum*, *Leishmania infantum*, and *Leptospira* spp. in dogs from the Fulni-ô indigenous community, located in Águas Belas, Pernambuco, Brazil. Blood samples were collected from 66 free-roaming or semi-domiciled dogs for serological analysis. Indirect immunofluorescence tests (IFAT) were used to detect antibodies for *T. gondii* and *N. caninum*, while *Leishmania infantum* detection was performed using the TR DPP® Canine Visceral Leishmaniasis kit. The presence of *Leptospira* spp. was evaluated for microscopic agglutination test (MAT). Results revealed seropositivity rates of 46.9% (31/66) for *T. gondii*, 9.1% (6/66) for *N. caninum*, 22.7% (15/66) for *Leishmania infantum*, and 18.2% (12/66) for *Leptospira* spp. Among the *Leptospira* spp. serovars identified, *Icterohaemorrhagiae* (83.3%, 10/12) and *Pomona* (50.0%, 6/12) were most prevalent, which were associated with environmental reservoirs and inadequate sanitation conditions. The findings suggest that the dogs in the Fulni-ô community act as environmental sentinels, reflecting local zoonotic risks. This pioneering study highlights the need for One Health-based strategies to prevent zoonotic diseases in vulnerable indigenous communities.

Keywords: public health, epidemiological surveillance, protozoa, neglected diseases.

Resumo

Doenças zoonóticas representam uma grande ameaça à saúde pública em comunidades indígenas devido ao isolamento geográfico, saneamento inadequado e contato próximo entre humanos, animais domésticos e animais selvagens. Este estudo teve como objetivo investigar a presença de anticorpos contra *Toxoplasma gondii*, *Neospora caninum*, *Leishmania infantum* e *Leptospira* spp. em cães da comunidade indígena Fulni-ô, localizada em Águas Belas, Pernambuco, Brasil. Amostras de sangue foram coletadas de 66 cães soltos ou semidomiciliados para análise sorológica. Testes de imunofluorescência indireta (RIFI) foram usados para detectar anticorpos para *T. gondii* e *N. caninum*, enquanto a detecção de *Leishmania infantum* foi realizada usando o kit TR DPP® Canine Visceral Leishmaniasis. A presença de *Leptospira* spp. foi avaliada por teste de aglutinação microscópica (MAT). Os resultados revelaram taxas de soropositividade de 46,9% (31/66) para *T. gondii*, 9,1% (6/66) para *N. caninum*, 22,7% (15/66) para *Leishmania infantum* e 18,2% (12/66) para *Leptospira* spp. Entre os sorovares de *Leptospira* spp. identificados, *Icterohaemorrhagiae* (83,3%, 10/12) e *Pomona* (50%, 6/12) foram os mais prevalentes, os quais foram associados a reservatórios ambientais e condições inadequadas de saneamento. Os resultados sugerem que os cães na comunidade Fulni-ô atuam como sentinelas ambientais, refletindo os riscos zoonóticos locais. Este estudo pioneiro destaca a necessidade de estratégias baseadas em Saúde Única para prevenir doenças zoonóticas em comunidades indígenas vulneráveis.

Palavras-chaves: saúde pública, vigilância epidemiológica, protozoários, doenças negligenciadas.

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1. Introduction

Emerging zoonotic diseases are estimated to account for approximately 2.4 billion cases and 2,7 million deaths annually worldwide (Harfield et al., 2015; Rahman et al., 2020). In Brazil, these diseases are particularly concerning in rural and semi-rural areas, such as indigenous communities, where close interactions between humans, domestic animals, and wildlife facilitate pathogen transmission (Canuto et al., 2018; Shah et al., 2024; Henrique et al., 2025).

The Fulni-ô Indigenous Territory, in Águas Belas, Pernambuco, is in the semi-arid Caatinga region and is home to approximately 4,000 people. Seasonality and recurrent droughts favor the persistence and spread of infectious agents, while isolation, the preservation of the Yathê language, and traditional practices restrict access to human and veterinary health services ((Bittencourt, 2022).

Indigenous communities represent one of the most vulnerable populations to zoonotic diseases due to geographic isolation, cultural barriers, and socioeconomic inequalities, which hinder access to both human and veterinary health services (Sousa et al., 2007; Baker et al., 2020). In these regions, the lack of veterinary services and proper management of dogs and other domestic animals contribute to their role as reservoirs for important zoonotic agents (Riley et al., 2022).

In Pernambuco and neighboring states, zoonoses such as visceral leishmaniasis, toxoplasmosis, neosporosis, and leptospirosis are endemic, with dogs serving as hosts or sentinels. Canine visceral leishmaniasis has been reported in several municipalities, as have leptospirosis outbreaks associated with heavy rainfall, reinforcing the need for canine surveillance in the semiarid Northeast region (Shapiro et al., 2019).

Canine visceral leishmaniasis has been reported in several municipalities of Pernambuco, while leptospirosis outbreaks have been documented during periods of heavy rainfall. These findings highlight the epidemiological importance of monitoring dog populations in semi-arid regions of Northeastern Brazil.

Moreover, the absence of robust surveillance systems hinders early detection of outbreaks, and the implementation of control strategies grounded in the One Health approach, which integrates human, animal, and environmental health (Riley et al., 2021).

Among the zoonotic agents associated with dogs are *Toxoplasma gondii*, *Neospora caninum*, *Leishmania infantum*, and *Leptospira* spp. These pathogens pose significant threats to public and animal health, along with economic implications (Pinto-Ferreira et al., 2019). In indigenous communities, dogs may act as environmental sentinels, reflecting the zoonotic risks faced by humans and other animals in these environments (Pinto-Ferreira et al., 2019; Riley et al., 2021, 2022, 2023).

Considering the historical marginalization of traditional peoples and the scarcity of studies on zoonoses in indigenous communities globally, this study aimed to investigate the presence of antibodies against *T. gondii*, *N. caninum*, *Leishmania infantum*, and *Leptospira* spp. in dogs from the Fulni-ô indigenous community, located in Pernambuco, Brazil.

2. Material and Methods

2.1. Study area and sampling

The Fulni-ô community is in the municipality of Águas Belas, within the Agreste region of Pernambuco, Brazil (Figure 1). The Fulni-ô people live on a reservation of approximately four-square leagues, administered by the National Indian Service (Hohenthal and McCorkle, 1955).

The geographic coordinates of the Fulniô reservation are approximately 9° 06′ 39″ S latitude and 37° 07′ 22″ W

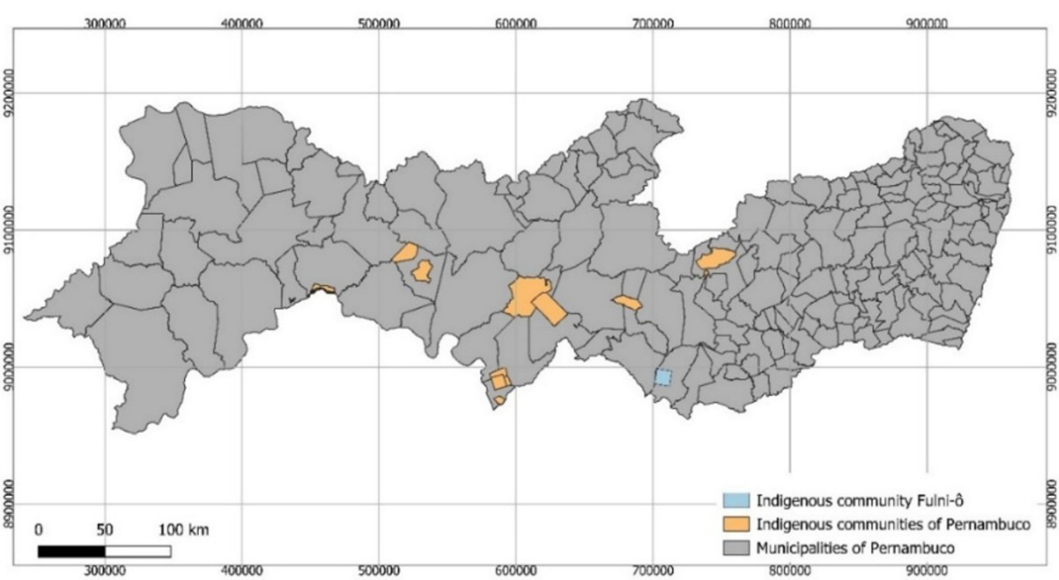

Figure 1. Location of the Fulni-ô indigenous community and other indigenous communities in Pernambuco state, Brazil.

longitude, situated at the foothills of the Serra do Cumanati range in the semi-arid agreste region of the São Francisco River basin, sub-basin of the Ipanema River (Silveira et al., 2012).

The local climate is characterized by low and irregular precipitation (500–700 mm/year) and average annual temperatures around 26 °C, conditions to which the community has adapted through sustainable water management practices and subsistence agriculture (Bittencourt, 2022).

Traditional Fulni-ô dwellings, originally built with straw and wood, are now complemented by modern masonry houses; the community's "ocras" (thatched structures) remain in use as sacred spaces for ancestral rituals (Bittencourt, 2022).

The residents maintain a bilingual culture (Portuguese and Yaathe) and practice a rich repertoire of ceremonies, including the "rasgadas" dances and the "ouricuri" rites, which promote social cohesion and are periodically open to visitors through the "cafurnas" cultural festival (Silveira et al., 2012).

Their livelihoods combine small-scale agriculture, artisanal extraction of medicinal plants (Albuquerque et al., 2011), and informal trade, while domestic animals roam freely or live in semi-domicile within the village limits, living in close contact with humans, other domestic animals, and local fauna (Santana; Magalhães, 2022).

Serum samples were collected from dogs of the Fulni-ô Indigenous community during a field campaign carried august 2018, as part of a One Health surveillance initiative in northeastern Brazil.

All 66 canine sera were obtained as convenience samples, corresponding to all free-ranging or semi-domiciled dogs encountered during field visits. The Inclusion criteria were age ≥ 3 months and owner consent (when domiciled). Due to logistical constraints and the absence of a formal canine census, we sampled all accessible animals that met these criteria, aiming to capture the broadest possible representation within the Fulni-ô reserve.

2.2. Ethical considerations

This study was approved by the Ethics Committee on the Use of Animals (CEUA) of the Universidade Federal Rural de Pernambuco (128/2017). This research was conducted as recommended by the Brazilian College of Animal Experimentation.

2.3. Biological samples

Blood samples were collected from free-ranging and semi-domiciled stray dogs of different breeds, sex, and ages by cephalic vein venipuncture, and stored in Vacutainer ® tubes (New Jersey, USA). The tubes were identified and centrifuged at 1,500 ×g for 5 min. to obtain blood serum. Sera were aliquoted into 1.5mL polypropylene tubes, identified, and stored at -20 °C until serological analysis.

Prior to blood collection, each dog underwent a brief physical examination, including measurement of rectal temperature, inspection of mucous membranes, palpation of peripheral lymph nodes, and assessment of skin and gait. No overt clinical signs (fever, lymphadenopathy, dermatologic or neurologic abnormalities) were detected in any of the 66 animals, which were therefore classified as clinically healthy at the time of sampling.

2.4. Serological analysis

2.4.1. Toxoplasma gondii and Neospora caninum

The detection of IgG antibodies against *T. gondii* and *N. caninum* was performed using the indirect immunofluorescence test (IFAT), with cut-off titters of 1:16 and 1:50, respectively (Dubey, 2010). Tachyzoites of the *T. gondii* strain ME-49 and the *N. caninum* strain Nc-Sp7 were used as antigens. Serum samples were diluted in phosphate-buffered saline (PBS) and applied to 12-well slides. A fluorescein-conjugated anti-canine IgG secondary antibody (Sigma Chemical®, USA) was added to each microwell containing the processed serum samples (Dubey, 2010). Positive and negative controls were included on each slide for both pathogens to ensure test validity.

2.4.2. Leishmania infantum

The presence of antibodies against *Leishmania infantum* was assessed using the TR DPP® 7 Canine Visceral Leishmaniasis Kit (Bio-Manguinhos, Brazil). The kit includes a DPP support, running buffer, disposable collection handle (5 µL capacity), and a disposable lancet. All procedures were performed following the manufacturer's instructions.

2.4.3. Leptospira spp.

The detection of antibodies against *Leptospira spp.* was carried out using the MAT, with a positivity threshold set at titters ≥ 50 (Bernardino et al., 2021a, b). A total of 15 serovars representing various serogroups were tested, as detailed in Table 1 (OIE, 2014).

The reaction was considered positive when titters equal to or greater than 1:50 were observed. The tiers obtained were classified into three categories: low (≤100), medium (200 to 400), and high (≥800). In addition, the mean titers observed for each serovar, and the frequency of different titer levels (low, medium, and high) were determined. A detailed analysis of co-positivity was also performed, identifying how many animals showed positivity for one or more serovars simultaneously.

2.5. Statistical analysis

Epidemiological and clinical information was obtained through a structured questionnaire applied to the owners or caretakers of the animals.

All statistical analyses were performed using IBM SPSS Statistics, version 26.0 (IBM Corp., Armonk, NY, USA). Associations between categorical variables (e.g., domicile status, contact with other animals, feeding type, and age category) and seropositivity to each pathogen were assessed using Pearson's chi-square test or Fisher's exact test when expected frequencies were <5. Statistical significance was set at $p < 0.05$ for all comparisons. (Thrusfield, 2007).

To identify possible associations between the different etiological agents investigated, Spearman's correlation test was used. The results of the analyses were organized into tables to facilitate visualization, interpretation, and presentation of the information obtained in this study.

3. Results

In the serological analysis of canine sera from the Fulniô community, the following antibody prevalences were observed: 46.9% (31/66) were reactive for *Toxoplasma gondii*; 9.1% (6/66) for *Neospora caninum*; 22.7% (15/66) for *Leishmania infantum*; and 18.2% (12/66) for *Leptospira* spp. (Table 1).

Regarding the overall serostatus, 28.8% (19/66) of dogs were seronegative for all tested agents. Most dogs, 60.9% (42/66), were reactive to only one pathogen. Coreactivity to more than one agent occurred in 11.6% (8/66) of animals, seven dogs (10.6%, 7/66) were positive for two pathogens and one dog (1.5%, 1/66) for three pathogens, while none reacted to all four agents. The most frequent dual reactivity was *T. gondii* + *Leptospira* spp. (10.6% 7/66), and the sole triple reactivity involved *T. gondii*, *Leptospira* spp. and *L. infantum* (Table 1).

In the characterization of epidemiological variables among the study population, most dogs (23/66) reported contact with all categories of animals (domestic, synanthropic and wild), while 15 dogs had no contact with other animals. Regarding domicile status, 47 dogs were domiciled, 12 semidomiciled and 9 nondomiciled. In terms of feeding, 36 dogs received a mixed diet of homecooked food plus commercial feed, 24 were fed exclusively homecooked food and 7 received only commercial feed. Age distribution peaked at 3 years (15 dogs), followed by 2 years (13 dogs) and dogs older than 10 years (10 animals).

An individual riskfactor analysis showed that “contact with other animals” was not significantly associated with any of the diseases investigated (toxoplasmosis p = 0.224; neosporosis p = 1.000; leptospirosis p = 0.965; leishmaniasis p = 0.392). Likewise, “type of feeding” showed no significant association with any of the infectious agents evaluated (*T. gondii* p = 0.545; *N. caninum* p = 1.000; *L. infantum* p = 0.841; *Leptospira* spp. p = 0.820).

Age category also had no statistically significant influence on disease occurrence, with all pvalues above 0.05 (*T. gondii* p = 0.511; *N. caninum* p = 1.000; *Leptospira* spp. p = 0.576; *L. infantum* p = 0.224). In contrast, domicile status was significantly associated with two pathogens: *L. infantum* (χ^2 = 9.22; p = 0.027) and *Leptospira* spp. (χ^2 = 8.97; p = 0.030), whereas no association was found for *T. gondii* (p = 0.606) or *N. caninum* (p = 1.000).

Of the 66 dogs evaluated, 12 (18.2%) were reactive by MAT for *Leptospira* spp. Among these, the serovar *Icterohaemorrhagiae* (ICT) was most frequent (83.3%, 10/12), followed by *Pomona* (POM; 50.0%, 6/12) and *Canicola* (CAN; 33.3%, 4/12). Each of the other serovars, *Hardjoprajitno* (HP), *Djasiman* (DJA), *Australis* (AUS), *Tarassovi* (TAR), *Grippotyphosa* (GRI) and *Bataviae* (BAT), was detected in one animal (8.3%, 1/12 each) (Table 2).

The highest titer observed among reactive sera was 1:1 600 for ICT, followed by 1:800 for POM, 1:400 for TAR, 1:200 for CAN, DJA and AUS, 1:100 for HP, and 1:50 for GRI and BAT. Overall, low titers (≤ 1:100) predominated. Only ICT exhibited reactivity across all titer categories (low, medium and high), whereas DJA, AUS and TAR were exclusively mediumtiter (1:200–1:400) and elevated titers (≥ 1:800) were observed only for ICT and POM.

With respect to coreactivity, seven dogs (10.6%) were positive for two serovars, two dogs (3.0%) for three serovars, and one dog (1.5%) for four distinct serovars. Only two dogs showed reactivity to a single serovar. Dog no. 3 was the sole animal reactive to four serovars (HP, CAN, GRI and BAT). Dogs no. 7 and no. 19 were reactive to three serovars each: dog 7 to DJA, AUS and TAR, and dog 19 to ICT, CAN and POM. Of the seven dogs with dual reactivity, nos. 5, 44, 54, 58 and 63 were positive for ICT + POM, while nos. 47 and 48 were positive for ICT + CAN. Dogs no. 28 and no. 62 were the only animals reactive to a single serovar both to ICT, which was the most frequently detected serovar in this sample.

Table 1. Seroprevalence of zoonotic agents detected in dogs from the Fulni-ô Indigenous community, Pernambuco, Brazil.

Pathogen / Disease	No. positive / total (%)	Diagnostic test	Main serovars or notes	Observed coinfections
Toxoplasma gondii	31 / 66 (46.9%)	IFAT	—	Most frequent dual reactivity: *T. gondii + Leptospira spp.*
Neospora caninum	6 / 66 (9.1%)	IFAT	—	No significant coinfection observed
Leishmania infantum	15 / 66 (22.7%)	TR DPP® Canine Visceral Leishmaniasis kit	—	One triple-positive dog (*T. gondii + Leptospira spp. + L. infantum*)
Leptospira spp.	12 / 66 (18.2%)	MAT	Predominant serovars: Icterohaemorrhagiae (83.3%), Pomona (50.0%), Canicola (33.3%)	7 dogs (10.6%) positive for *T. gondii + Leptospira spp.*
Overall serostatus	47 / 66 (71.2%) reactive to ≥1 pathogen	—	—	8 dogs (11.6%) reactive to ≥2 pathogens

Legend: IFAT: indirect fluorescent antibody test; MAT: microscopic agglutination test. Data represent antibody detection in serum samples collected from 66 dogs in the Fulni-ô Indigenous community, Águas Belas, Pernambuco, Brazil.

Table 2. Abbreviation of serovars and respective serogroups and species used in the MAT technique.

Sorovar	n	Percentage (%)
Icterohaemorrhagiae	10	83.3
Pomona	6	50
Canicola	4	33.3
Hardjo-prajitno	1	8.3
Djasiman	1	8.3
Australis	1	8.3
Tarassovi	1	8.3
Grippotyphosa	1	8.3
Bataviae	1	8.3

4. Discussion

The inclusion of domestic and production animals in serological investigations within indigenous communities is crucial, as these populations often rely on subsistence agriculture as their primary source of income and food. In this context, dogs and cats play vital roles, including pest management, property guarding, and companionship (Oliveira et al., 2011; Pulina et al., 2018).

Among the Kamentsá (Putumayo, Colombia), the canine seroprevalence of leptospirosis (80% by MAT) suggests intense environmental transmission and sanitation failures (H. Romero et al., 2018). In Manaus (urban), dogs have lower seroprevalence for *Toxoplasma gondii* (12.3%) and *Neospora caninum* (1.9%), indicating mitigation through infrastructure and management (Campos et al., 2022). In the rural Northeast (Paraíba), intermediate patterns emerge with co-exposure to Leptospira, *T. gondii*, and *N. caninum*, driven by household and wandering factors (Fernandes et al., 2018a). This contextualizes the Fulni-ô ecology, sanitation, and management determine risk.

However, these animals can also serve as reservoirs for zoonotic pathogens, contributing to the transmission of infectious diseases. The vulnerability of indigenous populations is exacerbated by poor sanitary conditions and close contact between humans, domestic animals, and wildlife, which facilitates the spread of infectious agents (Pulina et al., 2018; Romero-Sandoval et al., 2019).

The findings in dogs from the Fulni-ô community suggest that *Toxoplasma gondii* is circulating in the local environment. Infection with *T. gondii* occurs through the ingestion of infective oocysts in contaminated food or water, or through the consumption of raw meat from farm animals or wild prey (Innes et al., 2019; Awobode et al., 2020).

Indigenous contexts can also concentrate *T. gondii* exposure through food and landscape pathways. In Amazonian Indigenous communities, the handling and consumption of wild meat/offal have been linked to sustaining sylvatic *T. gondii* cycles. Proximity to forest edges and river margins has shown a spatial association with canine exposure in basin-wide studies (Fornazari et al., 2023). These determinants differ from predominantly urban exposures seen in Manaus, where contact with other dogs emerged as a key risk factor, highlighting how local subsistence practices and landscape shape infection ecology (Campos et al., 2022).

Studies conducted in indigenous communities in Brazil have shown that infection with *T. gondii* is associated with the consumption of water from contaminated rivers, the presence of felines, and cultural habits such as consuming raw mushrooms (Boia et al., 2008; Santos et al., 2019).

The presence of stray dogs in the village is particularly concerning, as these animals may play a role in the parasite's life cycle, potentially contaminating water and food consumed by humans and other intermediate hosts (Chalkowski et al., 2019; Arruda et al., 2021). Stray dogs act as indicators of environmental contamination (Opsteegh et al., 2019), underscoring the need for investigations in humans to estimate antibody prevalence and the health impacts of toxoplasmosis on the Fulni-ô population.

The detection of anti-*Neospora caninum* antibodies in dogs indicates an active transmission cycle of the protozoan. The presence of stray and semi-domiciled dogs in the community promotes the spread of *N. caninum*, especially given their propensity to hunt or consume raw meat—factors that have been associated with a higher risk of infection in rural areas (Sicupira et al., 2012). Previous studies have reported antibody prevalences in dogs from traditional communities, including 20.6% in semi-domiciled dogs in Australia (King et al., 2012) and 9.8% in indigenous villages in the Brazilian Amazon (Minervino et al., 2012).

In Brazil, anti-*N. caninum* antibodies have been detected in various domestic and wild animal species (Vianna et al., 2005; Gondim et al., 2010), reinforcing the role of dogs in maintaining the protozoan's transmission cycle. Moreover, *N. caninum* infection is a significant cause of abortion in cattle, particularly in regions where the disease is neglected (Abreu et al., 2014; Pinto-Ferreira et al., 2019).

Coinfections and diagnostic context warrant careful interpretation. In community surveys and necropsy-based series, co-seropositivity to protozoa is frequent, and clinical canine leishmaniasis has been associated with increased susceptibility to *T. gondii* in co-infected dogs, potentially modifying disease expression (Fornazari et al., 2023)

Moreover, serology panels (IFAT/ELISA) and MAT capture exposure but not necessarily active infection; cross-reactivity may occur, and confirmatory molecular testing adds clarity where feasible (Gaio et al., 2014; Fernandes et al., 2018b).

These caveats are relevant to Fulni-ô co-reactivity patterns (*T. gondii* and *Leptospira* spp.), which could reflect true co-exposure, shared environmental risks, or serologic cross-reaction.

The detection of antibodies against *Leishmania infantum* in dogs from the Fulni-ô community is noteworthy, especially given that the municipality of Águas Belas (PE) is classified as an endemic area for visceral leishmaniasis (VL). According to DataSUS/SINAN data, 29 cases of VL in humans were reported in the municipality between 2010 and 2024, indicating active circulation of the protozoan in the region. Additionally, one case of American tegumentary leishmaniasis (ATL) was recorded, further evidencing the disease's presence in the area (Brasil, 2025a, b).

Regionally, dog exposure profiles vary widely and frame the Fulni-ô signal. In the Xingu River Basin (Brazilian Amazon), dogs showed measurable exposure to *L. infantum* and *T. gondii* with spatial patterns linked to land cover, while a clinical cohort in Brazil's Northeast documented high Leishmania seropositivity in symptomatic dogs, emphasizing that the sampling frame (clinical vs. community) inflates apparent prevalence (Fornazari et al., 2023). Against this backdrop, the Fulni-ô community's 22.7% for *L. infantum* and 46.9% for *T. gondii* indicate substantial zoonotic circulation consistent with semi-arid ecology and close human–dog–wildlife interfaces.

The identification of seropositive dogs for *Leishmania infantum* suggests an ongoing risk of active transmission. However, the absence of confirmatory tests, such as PCR, limits the interpretation of these findings (Solano-Gallego et al., 2001). Integrated strategies, including the management of infected dogs, vector control, and health education, are essential. Prioritizing epidemiological characterization of leishmaniasis in the region is critical to protecting the Fulni-ô community and the broader population.

Leptospirosis, a widespread bacterial zoonosis, was also detected in dogs from the Fulni-ô community. The presence of various serovars reflects environmental reservoirs, such as synanthropic rodents (Cave et al., 2014; Lau et al., 2017). The natural infection of unvaccinated dogs likely occurred through exposure to contaminated water or moist soil, environments where leptospires can survive for extended periods (Thibeaux et al., 2017).

The serovar *Icterohaemorrhagiae* was the most prevalent (83.3%, 10/12) and is associated with high virulence, often resulting in severe cases of leptospirosis in humans and animals. This serovar is frequently linked to the presence of rats, its primary reservoirs, posing a significant risk in communities with poor sanitary conditions, such as indigenous communities (Samir et al., 2015; Riley et al., 2022).

The serovar *Pomona* (50%, 6/12) is highly pathogenic and frequently associated with pigs, though it is capable of infecting dogs and other species. Its detection in community dogs suggests environmental exposure or contact with sources of infection, such as contaminated water (Browne et al., 2022; Fahimipour et al., 2021).

The serovar *Canicola* (33.3%, 4/12), commonly found in dogs in Brazil, is linked to severe cases of renal failure. Dogs are the primary reservoirs of this serovar, and their shedding of bacteria through urine perpetuates transmission, posing a significant environmental risk (Hagiwara et al., 2015; Pellizzaro et al., 2019).

Other detected serovars, including *Hardjoprajitno*, *Dijasiman*, *Australis*, *Tarassovi*, *Grippotyphosa*, and *Bataviae* (8.3%, 1/12), highlight the diversity of *Leptospira* serovars circulating in the region. These are often associated with wild reservoirs and humid environments, which are common in rural areas (Thibeaux et al., 2017).

Detecting multiple serovars suggests repeated exposure or diverse sources of infection, characteristics of regions where frequent interactions occur between dogs, humans, and wild reservoirs. This underscores the complex epidemiological dynamics of leptospirosis in rural and semi-rural indigenous communities.

Actionable One Health implications follow from these patterns. In Indigenous territories with high Leptospira serogroup diversity and relevant *T. gondii* and *L. infantum* exposure, targeted steps include environmental sanitation, rodent control, safe water access, and health education on handling/cooking wild meat, alongside dog management (roaming reduction), vaccination where applicable, and periodic dog-sentinel surveillance (Campos et al., 2022; Galvão et al., 2023a, b; Khan et al., 2023).

Evidence from Indigenous and rural settings shows that sanitation and husbandry improvements correspond with lower dog seroprevalence, supporting integrated interventions with veterinary services embedded in Indigenous health systems (H. Romero et al., 2018).

We acknowledge that the limited sample size and convenience sampling approach limit the generalizability of our findings, but the detection of multiple pathogens and zoonotic agents in this population highlights the complex epidemiological dynamics at the rural-urban interface of indigenous communities, reflecting repeated exposure or multiple sources of infection caused by frequent interactions between dogs, humans, and wildlife.

5. Conclusion

Dogs in the Fulni-ô community showed extensive exposure to *Toxoplasma gondii*, *Neospora caninum*, *Leishmania infantum*, and *Leptospira* serovars, indicating an active zoonotic risk at the human-dog-environment interface. From a One Health perspective, integrated and periodic surveillance, environmental sanitation and vector control actions, and measures aimed at dogs such as reducing straying, vaccination, and health education are recommended. The proposed protocol can be replicated in other Indigenous territories, adjusted to local ecology and customs, serving as a model to strengthen zoonosis surveillance and prevention nationwide.

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Data Availability Statement

The datasets generated and analyzed during the current study are not publicly available due to ethical and legal restrictions related to the conservation status and handling of wildlife species. However, anonymized data supporting the findings of this study are available from the corresponding author upon reasonable request.

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