



Detection of anti-*Toxoplasma gondii* antibodies in *Callithrix* spp. in Espírito Santo, Brazil¹

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ABSTRACT.- Freitas MRB, Carnieli JS, Carmo ST, Athayde-Gusmão AE, Silva AF, Raimundo JM, Baldani CD, Guimarães A. **Detection of anti-*Toxoplasma gondii* antibodies in *Callithrix* spp. in Espírito Santo, Brazil.** *Pesquisa Veterinária Brasileira* 46:e07806, 2026. Instituto de Veterinária, Universidade Federal Rural do Rio de Janeiro, Rodovia BR-465 Km 07, Zona Rural, Seropédica, RJ 23890-000, Brazil. E-mail: andresaguimaraes02@yahoo.com.br

Toxoplasmosis is an infectious disease caused by the obligate intracellular protozoan *Toxoplasma gondii*, which has a worldwide distribution and is recognized for frequently causing acute and fatal disease in neotropical primates. This study aimed to detect anti-*T. gondii* antibodies in *Callithrix* spp. from different locations in Espírito Santo, Brazil, and to describe the distribution of seroreactivity according to environmental and host-related variables. A total of 21 animals were evaluated from 2022 to 2025, both sexes and ranging in age from juveniles to adults, including individuals maintained under human care and free-ranging individuals. Seroreactivity was detected using the modified agglutination test in 23.8% (5/21) of the primates, with titers of 1:25 and 1:50. All seroreactive individuals were identified among those classified under human care at the time of sampling, while no reactivity was observed among free-ranging individuals. The distribution of seroreactivity did not suggest a clear pattern in relation to sex or age and should be interpreted with caution due to the structure of the sampled population. The detected pattern was associated with the environmental and management contexts of the municipalities of origin of the animals, with seroreactive individuals linked to areas with greater anthropogenic influence. The low antibody titers observed are consistent with previous reports involving neotropical primates; however, the exclusive use of a serological method and the high susceptibility of this group may limit the detection of infection, particularly in cases of acute disease progression. These findings provide insights into *T. gondii* exposure in neotropical primates from Espírito Santo and highlight the importance of integrating serological and molecular approaches in future studies to better characterize parasite circulation and epidemiological dynamics in the region.

INDEX TERMS: Toxoplasmosis, neotropical primates, serology, environment.

RESUMO.- [Detecção de anticorpos anti-*Toxoplasma gondii* em primatas do gênero *Callithrix* no Espírito Santo, Brasil.] A toxoplasmose é uma doença infecciosa causada pelo protozoário intracelular obrigatório *Toxoplasma gondii*, de distribuição mundial, reconhecido por causar com frequência condição aguda e fatal em primatas neotropicals. Este estudo

buscou como objetivo detectar anticorpos anti-*T.gondii* em *Callithrix* spp. provenientes de diferentes locais do Espírito Santo, Brasil, e descrever a distribuição de sororreatividade de acordo com variáveis ambientais e relacionadas ao hospedeiro. Foram avaliados 21 animais, de 2022 a 2025, de ambos os sexos e com faixa etária variando de juvenil a adulto, sendo 10 mantidos sob cuidados humanos e 11 de vida livre. Foi detectada sororreatividade através do teste de aglutinação modificado em 23,8% (5/21) dos primatas, com titulações de 1:25 e 1:50. Todos os indivíduos sororreagentes foram identificados entre aqueles classificados como mantidos sob cuidados humanos no momento da coleta, enquanto nenhuma reatividade foi observada entre os indivíduos de vida livre.

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A distribuição da sororreatividade não sugeriu um padrão claro em relação ao sexo ou à idade e deve ser interpretada com cautela devido à estrutura da população amostrada. O padrão observado foi associado aos contextos ambientais e de manejo dos municípios de origem dos animais, com indivíduos sororreagentes associados a áreas com maior influência antrópica. As baixas titulações relatadas são consistentes com a literatura, no entanto, o uso exclusivo de um método sorológico e a alta suscetibilidade desse grupo podem limitar a detecção da infecção, especialmente em casos de progressão aguda da doença. Esses achados fornecem informações relevantes sobre a exposição ao *T. gondii* em primatas neotropicais do Espírito Santo e destacam a importância da integração de abordagens sorológicas e moleculares em estudos futuros, visando melhor caracterizar a circulação do parasito e a dinâmica epidemiológica na região.

TERMOS DE INDEXAÇÃO: Toxoplasmose, primatas neotropicais, sorologia, meio ambiente.

INTRODUCTION

Toxoplasma gondii is an obligate intracellular protozoan with worldwide distribution that affects a wide range of animals, including wild and domestic felids, which are considered definitive hosts and shed oocysts in their feces. Other homeothermic animals, including non-human primates, are considered intermediate hosts in which tissue cysts are formed (Dubey 2004). Toxoplasmosis may present in acute or chronic forms, varying according to factors such as the strain involved, its degree of pathogenicity, and the interaction between the parasite, the host, and the immunological status of the affected individual (Paula et al. 2020).

The life cycle of *T. gondii* is heteroxenous and requires at least two hosts for its maintenance. In intermediate hosts, asexual reproduction occurs and involves two parasite forms: tachyzoites, which show rapid multiplication and dissemination throughout the organism during the acute phase of infection, and bradyzoites, which form tissue cysts during the chronic phase (Dubey 2004). In definitive hosts, sexual reproduction occurs in the intestine, producing oocysts that are shed in feces, sporulate in the environment, and become infective. This diversity of forms accounts for the parasite's high transmission efficiency and ecological persistence, with infection in humans and animals occurring through ingestion of oocysts present in water, soil, or contaminated food, or through the consumption of raw or undercooked meat containing tissue cysts (Tenter et al. 2000).

The relationship between non-human primates and toxoplasmosis varies according to species, with immunological, behavioral, and ecological factors playing important roles. New World primates, such as *Callithrix* spp. (marmosets), are clinically more susceptible to infection, showing acute clinical signs and higher fatality rates compared with Old World primates (Minervino et al. 2017, Dubey et al. 2021). In New World primates, the clinical presentation is usually severe and multisystemic, with signs such as apathy, anorexia, abdominal distension, and fever (Casagrande et al. 2013), and lesions compatible with acute toxoplasmosis, including necrotizing hepatitis, interstitial pneumonia, and neutrophilic encephalitis, which are widely described in cases of sudden death (Casagrande et al. 2013, Paula et al. 2020). However,

among New World primate species, *Sapajus* spp. (capuchin monkeys) often show higher seropositivity without compatible lesions, suggesting possible adaptation and greater exposure to the parasite (Gusmão 2024).

In addition to the inherent differences in susceptibility between Old World and New World primates, environmental factors may influence the probability of exposure. In free-ranging animals, space use, diet, and the degree of contact with areas under human influence affect the risk of contact with oocysts, potentially favoring higher exposure in species with more terrestrial habits, such as *Cebus* spp. and *Sapajus* spp., compared with more arboreal species, such as *Callithrix* spp. (Niehaus et al. 2020, Gusmão 2024). Primates maintained under human care may be subject to different exposure conditions related to management practices, closer proximity to domestic felids, and the hygienic conditions of enclosures (Pires et al. 2012, Minervino et al. 2017, Gusmão 2024).

Serological methods are considered the main tools for the assessment of exposure to *T. gondii* in wildlife. The modified agglutination test (MAT) is commonly applied in non-human primates due to high specificity, independence from species-specific conjugates and ability to detect low antibody titers (Dubey & Desmonts 1987, Minervino et al. 2017, Niehaus et al. 2020, Carvalho et al. 2025). Although serological assays reflect previous exposure rather than active infection, they do not allow the identification of circulating genotypes (Dubey et al. 2021). Furthermore, serology by itself may fail to detect acute fatal infections when used in assays with highly susceptible species, as infected individuals may die before seroconversion, reinforcing the importance of integration with molecular diagnostic methods and cautious interpretation of results (Casagrande et al. 2013, Oliveira et al. 2022, Zorzi et al. 2025).

Espírito Santo lacks studies addressing the health of primate fauna. The state hosts wildlife screening and rehabilitation centers that assist in the treatment, maintenance, and rehabilitation of animals rescued from accidents, illegal trade, and other anthropogenic impacts. Conversely, several protected areas and reserves harbor high faunal diversity within Atlantic Forest fragments. In both contexts, the presence of *Callithrix* spp. is consistent; therefore, investigating the pathogens to which these primates are exposed is of great importance, particularly considering the zoonotic potential of toxoplasmosis. Given the combined influence of environmental and management-related factors on exposure to oocysts, it is important to investigate the occurrence of *T. gondii* exposure in Neotropical primates from different contexts. Accordingly, the present study aimed to detect anti-*T. gondii* antibodies in *Callithrix* spp. from different localities in Espírito Santo, Brazil, and to describe the distribution of seroreactivity according to environmental and host-related variables, providing insights into exposure in neotropical primates from the region.

MATERIALS AND METHODS

Ethical approval. This study was approved by the Ethics Committee on the Use of Animals (CEUA) of the "Instituto Federal do Espírito Santo" (IFES), under process number 23152.000419/2022-47 and authorization number from the "Sistema de Autorização e Informação em Biodiversidade" (Biodiversity Authorization and Information System – SISBIO) 71888.

Animals evaluated. A total of 21 clinically healthy neotropical primates, both males and females, from the municipalities of

Santa Teresa, Serra, and Aracruz, in the state of Espírito Santo, Brazil, were included in this study from 2022 to 2025. Regarding age classification, infants were excluded, and only juveniles and adults were sampled. Age category was determined based on body weight, size, and dentition, following criteria adopted for other primate species (Eadie 2015). All sampled primates belonged to the genus *Callithrix*, with 10 identified as *Callithrix geoffroyi* and 11 as *Callithrix* sp. (hybrids). Of these, 11 were free-ranging, and 10 were maintained under human care. Free-ranging individuals originated from the “Instituto Nacional da Mata Atlântica” (National Institute of the Atlantic Forest - INMA; n = 5), the “Estação Biológica de Santa Lúcia” (Santa Lúcia Biological Station - EBSL; n = 3), and the Macaco Barbado “Reserva Particular do Patrimônio Natural” (Private Natural Heritage Reserve - RPPN; n = 3), all located in the municipality of Santa Teresa. At INMA and the RPPN, all primates were identified as *Callithrix* sp., whereas at EBSL, two individuals were identified as *Callithrix* sp. and one as *C. geoffroyi*. Primates maintained under human care were obtained from the Espírito Santo “Centro de Triagem de Animais Silvestres” (Wildlife Screening Center - CETAS; n = 5), located in the municipality of Serra, and from the “Centro de Reintrodução de Animais Selvagens” (Wildlife Reintroduction Center - CERÉIAS; n = 5), located in Aracruz. All individuals from CERÉIAS were identified as *C. geoffroyi*, whereas at CETAS, four *C. geoffroyi* and one *Callithrix* sp. were evaluated. In this study, animals classified as “maintained under human care” were those housed at CETAS or CERÉIAS at the time of sampling, irrespective of their prior origin. As detailed records of the duration of stay were unavailable, it was not possible to differentiate recent admissions from long-term residents. Therefore, this classification reflects the management and environmental conditions at the time of sampling rather than long-term captivity status. This limitation was considered in data interpretation and highlights the need for cautious interpretation of findings related to different management and environmental contexts.

Sample collection. All animals were subjected to chemical restraint to allow a safe clinical evaluation. The anesthetic protocol consisted of an association of ketamine (1.5 mg/kg) and midazolam (0.5 mg/kg), administered intramuscularly into the quadriceps muscle. Each individual was examined, and a clinical record was completed with information on estimated age, sex, physiological parameters (heart rate, respiratory rate, and rectal temperature), and medical history when available. For free-ranging animals, capture was performed using monitored Tomahawk traps. Animals were removed shortly after capture and subsequently placed in restraint bags for clinical procedures. Approximately 1 mL of blood was collected by femoral vein puncture and stored in tubes with a clot activator, which were centrifuged to obtain serum. Samples were kept refrigerated, then

placed in an insulated container and transported to the “Laboratório de Patologia Clínica Veterinária” (Veterinary Clinical Pathology Laboratory - LABVET) of the “Instituto de Veterinária” (Institute of Veterinary Medicine) of the “Universidade Federal Rural do Rio de Janeiro” (UFRRJ).

Laboratory processing for antibody detection. Using serum samples, the modified agglutination test (MAT) was performed at the Apicomplexa Research Laboratory of UFRRJ, according to the protocol previously established by Dubey & Desmonts (1987). A commercially obtained antigen solution was used, composed of formalin-fixed *Toxoplasma gondii* RH strain tachyzoites, borate buffer, Evans blue dye, and 2-β-mercaptoethanol. The antigen mixture (25 µL per well) was distributed into 96-well U-bottom microplates, followed by the addition of serum samples diluted 1:25 in phosphate-buffered saline (PBS), at a 1:1 ratio. Positive and negative control samples were also included in the assay. Plates were incubated at 37 °C for 12 hours. Samples showing a blue button at the bottom of the well were considered negative, whereas those with a clear well bottom were considered positive. Positive samples were subjected to serial dilution titration at 1:25, 1:50, 1:100, and 1:200, following the same protocol, to determine the final antibody titer for each reactive primate, with 1:25 established as the cutoff point. Reaction readings were performed by visual inspection.

Data analysis. Due to the uneven distribution of individuals across species and habitat conditions and the limited sample size, data were analyzed using a descriptive approach. Seroreactivity was described according to the categorical variables sex (male/female), age (juvenile/adult), species (*C. geoffroyi*/*Callithrix* sp.), and habitat condition (free-ranging/maintained under human care).

RESULTS

The distribution of individuals across species and habitat conditions was unbalanced, reflecting the opportunistic nature of wildlife sampling in rescue, rehabilitation and free-ranging contexts. Overall seroreactivity among the samples was 23.8% (5/21). All seroreactive individuals were identified as *Callithrix geoffroyi*. Among these, four originated from CERÉIAS and presented a titer of 1:25. At the same time, one individual from CETAS showed a titer of 1:50, the highest observed in the study, and no seroreactivity was detected among the remaining individuals. The distribution of seroreactivity according to sampling localities and antibody titers is presented in Table 1, while the distribution according to sex, age, species and habitat condition is shown in Table 2. The geographic distribution of the sampling sites is presented in Figure 1.

Table 1. Seroreactivity and antibody titers against *Toxoplasma gondii* determined by the modified agglutination test in neotropical primates from different localities in Espírito Santo, Brazil

Origin	Habitat condition	Species	Number of animals	Seroreactive (n)	Titers (n)
CETAS (Serra)	Under human care	<i>Callithrix geoffroyi</i> and <i>Callithrix</i> sp.	5	1 (20%)	1:50
CERÉIAS (Aracruz)	Under human care	<i>Callithrix geoffroyi</i>	5	4 (80%)	1:25
INMA (Santa Teresa)	Free-ranging	<i>Callithrix</i> sp.	5	0 (0%)	-
EBSL (Santa Teresa)	Free-ranging	<i>Callithrix geoffroyi</i> and <i>Callithrix</i> sp.	3	0 (0%)	-
RPPN (Santa Teresa)	Free-ranging	<i>Callithrix</i> sp.	3	0 (0%)	-
TOTAL	-	-	21	5	1:25 (n = 4) and 1:50 (n = 1)

CETAS = “Centro de Triagem de Animais Silvestres” (Wildlife Screening Center), CERÉIAS = “Centro de Reintrodução de Animais Selvagens” (Wildlife Reintroduction Center), INMA = “Instituto Nacional da Mata Atlântica” (National Institute of the Atlantic Forest), EBSL = “Estação Biológica de Santa Lúcia” (Santa Lúcia Biological Station), RPPN = “Reserva Particular do Patrimônio Natural” (Private Natural Heritage Reserve).

DISCUSSION

In the present study, 23.8% (5/21) of *Callithrix* spp. samples showed seroreactivity to *Toxoplasma gondii*, with seroreactive individuals exclusively identified among those classified under human care. It is important to note that animals were classified according to their condition at the time of sampling, regardless of their previous history, which does not allow differentiation between recent admissions and long-term maintenance. This pattern may be interpreted given the ecological and biological characteristics of the genus, such as predominantly arboreal habits, which may influence contact with the parasite. Furthermore, considering the high susceptibility of these primates, infections may progress rapidly in an acute and fatal manner, potentially reducing the likelihood of survival until seroconversion and, consequently, the detection of antibodies in serological surveys (Casagrande et al. 2013, Moura 2018, Paula et al. 2020). In addition, the distribution of seroreactivity according to species, age, and sex should also be interpreted with caution, as it may be influenced by the structure of the sampled population. Similar patterns of variation in serological detection have been reported in other studies involving neotropical primates, in which detection varied according to ecological context and host characteristics. At the same time, the influence of variables such as species, age, and sex remains inconsistent across studies (Molina et al. 2014, Niehaus et al. 2020, Gusmão 2024).

Seroreactivity detected in primates maintained under human care in the present study is consistent with other studies reporting higher detection under conditions of human care or temporary maintenance. Minervino et al. (2017), in a study conducted at the “Centro Nacional de Primatas” (National Primate Center) in Pará, reported a seroreactivity of 49.2%, suggesting that environmental conditions in enclosures may contribute to exposure, including the presence of oocysts. Niehaus et al. (2020) found that, among neotropical primates in Costa Rica, seroreactivity was 11.6% in free-ranging animals, whereas it reached 59.6% among those under human care. Complementarily, Silva et al. (2024) discuss that factors such as hygiene conditions of water, food, and enclosures represent relevant sources of risk and may favor environmental contamination. In this context, management-related factors, such as feeding practices, water supply systems, and enclosure

substrates, may represent potential sources of exposure and should be considered in the interpretation of the results.

In addition to factors directly related to management, other aspects of the evaluated population may also be considered when interpreting the observed pattern. Many of the seroreactive animals originated from a rehabilitation context, with possible prior exposure to environments influenced by human activities, including urban areas, even before entering the facilities. This situation was discussed by Pires et al. (2012), who emphasized that a large proportion of animals received by institutions such as CETAS have previously passed through anthropogenic areas. Furthermore, Niehaus et al. (2020) discuss that variables such as reduced forest cover and the presence of adjacent urban areas may influence exposure prior to the admission of individuals into enclosures. Therefore, the seroreactivity observed in primates classified as under human care may also reflect exposure before rescue, rather than being exclusively associated with institutional settings.

The results may also be interpreted in association with the environmental characteristics of the municipalities of origin of the primates. INMA, EBSL, and the Macaco Barbado RPPN are located in the municipality of Santa Teresa, which is known for harboring extensive remnants of the Atlantic Forest, with high forest connectivity and a consolidated history of conservation (Mendes & Padovan 2000). These locations are inserted in a context of greater environmental integrity,

Table 2. Distribution of seroreactivity to *Toxoplasma gondii* according to sex, age, species, and habitat condition in *Callithrix* primates in Espírito Santo, Brazil

Variables	Seroreactive (n)	Non-reactive (n)
Sex		
Male	3 (23.1%)	10 (76.9%)
Female	2 (25%)	6 (75%)
Age		
Juvenile	0 (0%)	5 (100%)
Adult	5 (31.3%)	11 (68.8%)
Species		
<i>Callithrix geoffroyi</i>	5 (50%)	5 (50%)
<i>Callithrix</i> sp.	0 (0%)	11 (100%)
Habitat condition		
Free-ranging	0 (0%)	11 (100%)
Under human care	5 (50%)	5 (50%)

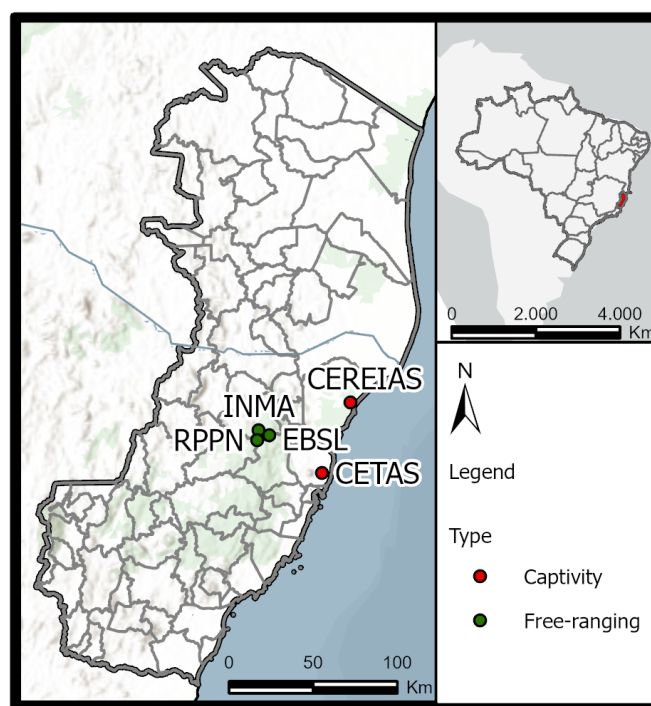


Fig. 1. Sampling sites of *Callithrix* spp. in Espírito Santo, Brazil. CETAS = “Centro de Triagem de Animais Silvestres” (Wildlife Screening Center), CERIAS = “Centro de Reintrodução de Animais Selvagens” (Wildlife Reintroduction Center), INMA = “Instituto Nacional da Mata Atlântica” (National Institute of the Atlantic Forest), EBSL = “Estação Biológica de Santa Lúcia” (Santa Lúcia Biological Station), RPPN = “Reserva Particular do Patrimônio Natural” (Private Natural Heritage Reserve).

which may be associated with reduced anthropogenic sources of contamination. They may contribute to the absence of seroreactivity among the evaluated individuals from these areas. The Macaco Barbado Reserve presents an important particularity, as it is located in a private area close to residential zones, yet no individuals from this site showed seroreactivity.

In contrast, the CETAS facility, located in the municipality of Serra, is situated in a region characterized by intense forest fragmentation and urban expansion, including the advance of industrial and residential areas, as reported by Lovate & Coelho (2025). Animals originating from CEREIAS, located in the municipality of Aracruz, are inserted in a comparable scenario, as the municipality has undergone significant urban transformations over recent decades, mainly associated with industrialization and population growth (Orrico 2010). In this context, environmental conditions associated with greater human influence may represent potential sources of exposure and should be considered when interpreting the results.

The distribution of seroreactivity according to sex and age in the present study did not suggest a clear pattern, consistent with previous studies involving neotropical primates that also did not identify consistent associations between exposure and these variables (De Moura et al. 2021, Silva et al. 2024). In a study conducted specifically with *Callithrix* spp., Oliveira et al. (2022) likewise reported no clear connection. Regarding habitat condition, seroreactivity was observed only among individuals classified as under human care, a pattern also described in studies reporting serological detection in animals under similar conditions (Pires et al. 2012, Minervino et al. 2017, Niehaus et al. 2020, Silva et al. 2024). The distribution of seroreactivity according to species should be interpreted with caution, as species and habitat conditions were overlapping variables in the present study. Most individuals considered under human care were identified as *Callithrix geoffroyi*, whereas *Callithrix* sp. individuals were predominantly considered free-ranging. Therefore, the observed pattern in relation to species may reflect the distribution of individuals across different environmental contexts rather than intrinsic species-specific susceptibility. Additionally, the limited sample size should be considered when interpreting these findings, reinforcing the descriptive nature of the analysis.

Antibody titers observed in this study were low, with four samples presenting a titer of 1:25 and only one reaching 1:50, with no reactivity at higher dilutions. This pattern has also been reported in other studies involving neotropical primates. In investigations conducted with *Callithrix* spp. under human care or temporary management conditions, titers are often discrete, as reported by Pires et al. (2012), who recorded a titer of 1:32 in *Callithrix* sp., and by Molina et al. (2014) and Gusmão (2024), who reported titers of 1:25 in free-ranging individuals. These findings suggest that, even under different contexts, *T. gondii* infections in these animals often result in low antibody titers. In contrast, in primates considered more resistant, such as *Sapajus* spp., higher titers occur more frequently, as shown in the study by De Moura et al. (2022), which detected a titer of 1:12,800 in *Sapajus apella*. Nevertheless, most individuals will present low titers regardless of species. The ability of MAT to detect both low and high titers was demonstrated in a comparative study by Carvalho et al. (2025), which showed greater sensitivity than indirect immunofluorescence by detecting a higher number

of seroreactive primates, including several with low titers, supporting its suitability as an effective screening method for primates.

Finally, interpretation of serological surveys in neotropical primates should consider the high susceptibility of this group as an important limitation. Although serology is the main tool for assessing exposure to *T. gondii*, in highly susceptible species, infections may progress to death before sufficient time elapses for the production of detectable antibodies. In such cases, infected individuals may remain seronegative, reducing the ability of serological surveys to capture the full extent of the parasite's presence within populations (Oliveira et al. 2022). The combination of serological and molecular methods can broaden the epidemiological understanding of toxoplasmosis, as each approach provides different and complementary information. While serology indicates previous exposure, DNA detection confirms the direct presence of the agent. Zorzi et al. (2025) demonstrated this complementarity by applying MAT and quantitative polymerase chain reaction (qPCR) in primates maintained under confinement, showing that the inclusion of molecular methods may help more comprehensively characterize infection in the evaluated populations. In the present study, antibody detection suggests subclinical exposure; however, it does not confirm active infection or characterize circulating parasite genotypes, aspects that may be further clarified by implementing molecular methods in future investigations.

CONCLUSIONS

Seroreactivity to *Toxoplasma gondii* was observed in part of the evaluated *Callithrix* spp., with distribution related to the management and environmental contexts of the animals' municipalities of origin. In particular, primates from areas with greater environmental integrity, such as Santa Teresa, where Atlantic Forest remnants are better preserved, showed no seroreactivity. In contrast, individuals from regions with more human influence, such as Serra and Aracruz, presented seroreactivity.

The detection of antibodies in primates from these localities provides relevant information on parasite circulation in different environmental contexts of Espírito Santo. It may contribute to further understanding the exposure patterns in neotropical primates from the region. These findings highlight the importance of considering local environmental characteristics, as well as factors related to previous exposure history and animal management, when interpreting serological data in wildlife.

No clear pattern of seroreactivity was observed associated with individual variables such as sex and age, which is consistent with previous reports. Nevertheless, the classification of animals based on their condition at the time of sampling and the lack of information regarding prior exposure limit the interpretation of the results. In addition, the small sample size and the exclusive use of serological methods reinforce the need for complementary studies incorporating environmental analyses and molecular approaches to better characterize the epidemiology of *T. gondii* in these populations.

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Conflict of interest statement.- The authors declare that there are no conflicts of interest.

Credit author statement.- Marcos R.B. Freitas - Writing and support in performing the modified agglutination test. Juliana de S. Carnieli - Sample processing and support in performing the modified agglutination test. Sarisha T. do Carmo - Animal capture, veterinary care and sample collection. Anna E. Athayde-Gusmão - Performing modified agglutination test. Andressa F. da Silva - Performing modified agglutination test. Cristiane D. Baldani - Sample processing. Andresa Guimarães - Conceptualization, data analysis, review, and supervision.

Data availability statement.- The data that support the findings of this study are available from the corresponding author upon reasonable request.

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