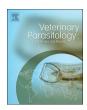
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Short Communication

Detection of anti-*Toxoplasma gondii* antibodies in small wild mammals from preserved and non-preserved areas in the Caatinga biome, a semi-arid region of Northeast Brazil



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ABSTRACT

This study was conducted to determine the seroprevalence of anti-*Toxoplasma gondii* antibodies in 152 free-living small wild mammals from distinct regions in the Caatinga biome, a semi-arid region in the Northeast of Brazil: the National Park of Serra das Confusões (NPSC), which is a preserved area in the state of Piauí, and the municipalities of Petrolina and Lagoa Grande, two non-preserved areas in the state of Pernambuco. Using the modified agglutination test (MAT), we found that 5.3% (4/75) and 3.3% (2/60) of small wild mammals were positive for IgG anti-*T. gondii* antibodies in the NPSC and Petrolina, respectively. All mammals from Lagoa Grande (0/17) tested negative on the MAT. Indirect infection of *T. gondii* was determined by MAT in *Galea spixii*, *Monodelphis domestica* and *Thrichomys laurentius* (from NPSC) and in *Didelphis albiventris* (from Petrolina). Seropositive animals were observed in both preserved and non-preserved areas within the Caatinga biome. Low seroprevalences observed can be related to the extreme temperature and humidity in this particular biome.

1. Introduction

Toxoplasma gondii, an intracellular parasite belonging to the phylum Apicomplexa, has a facultative heteroxenous life cycle and can infect all warm-blooded animals. The protozoan is globally prevalent and highly impactful in veterinary medicine and public health. Toxoplasmosis is a zoonosis that can cause abortion and congenital diseases in intermediate hosts (Tenter et al., 2000). Domestic and wild cats are the definitive hosts for *T. gondii* and are the only hosts that can excrete resistant oocysts in their faeces. Warm-blooded animals are infected mainly by ingesting the oocysts, which may be present as contaminants in the water or in the environment in general, and by ingesting cysts containing bradyzoites through consumption of tissues from infected hosts (Dubey, 2010).

The Caatinga biome is located in a semi-arid region of Brazil. The main characteristic of the semi-arid region is its low rainfall, which is, on average, $<600\,\mathrm{mm}$ per year. The rainy season is restricted to three

or four months of the year, while the dry season can encompass 70% of the year (Marengo et al., 2011). The region experiences intense sun on almost every day of the year, and the average temperature varies between 23 °C and 27 °C (Teixeira, 2016). The environmental conditions of the Caatinga sharply contrast with those of the other Brazilian biomes; they include rainfall irregularity from year to year, which results in severe droughts and an inhospitable environment for local fauna (Krol et al., 2001). Preserved and non-preserved areas can be found in this biome. A preserved area is an area of environmental protection and has almost no human activity; thus, greater biodiversity is expected among species. Non-preserved areas, on the other hand, have experienced constant human activity over the decades in the urban and rural environments, resulting in more intense environmental degradation and lower biodiversity.

The present study aimed to evaluate the prevalence of anti-*T. gondii* antibodies in free-living small wild mammals from three distinct areas (one preserved and two non-preserved) within the Caatinga biome of

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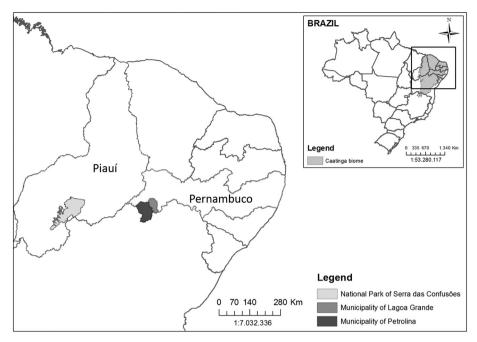


Fig. 1. Map of Northeast Brazil with the locations from where animals were sampled in the states of Piauí and Pernambuco.

Northeast Brazil.

2. Materials and methods

2.1. Study area

This study was conducted in distinct areas within the Caatinga biome in the semi-arid region of Northeast Brazil: the National Park of Serra das Confusões (NPSC), a preserved area that is in the southeastern part of the state of Piauí (9° 00′ 57″ S, 43° 26′ 51″ W), and the municipalities of Petrolina (9° 23′ 55″ S, 40° 30′ 03″ W) and Lagoa Grande (9° 19′ 42″ S, 40° 33′ 17″ W), both non-preserved areas that are in the western part of the state of Pernambuco (Fig. 1). This biome, a mosaic of thorny xerophilous bushes and forest that covers much of Northeast Brazil and part of the state of Minas Gerais, extends 735,000 km². The NPSC is one of the largest and most recent conservation units of the Caatinga biome and covers a total area of 823,435 ha. The municipalities of Petrolina and Lagoa Grande are located in the San Francisco Valley region, representing medium-sized municipalities within an environment that is degraded due to intense human activity.

2.2. Capture and collection of blood from wild mammals

We made four visits to different areas in the NPSC from January to October 2013, four visits to the municipality of Petrolina from August 2014 to April 2015, and four visits to the municipality of Lagoa Grande from August 2014 to April 2015. Each area was visited in only one period in each season of the year. These years of sample collection were part of a five-year period (2011-2016) of the most severe, prolonged and extensive drought in the semi-arid Northeast Brazil region (Brito et al., 2018). The wild mammal capture sites were selected according to the feasibility of trap placement and the availability of vegetation. In the NPSC, Petrolina and Lagoa Grande, a total of 70, 92 and 92 traps were used each night, respectively, for four nights per visit. Marsupials and rodents were captured using Tomahawk (45 \times 16 \times 16 cm) and/or Sherman (30 \times 8 \times 9 cm and 43 \times 12,5 \times 14,5 cm) traps, which were baited with grits, sardines, bagels, bananas and pineapples and placed in shaded areas near trees. Initially, physical restraint was applied with the help of gloves, and then weighing and subsequent chemical restraint were applied using 15-30 mg/kg of ketamine hydrochloride (Cubas et al., 2014). Blood collection from marsupials and rodents was performed by intra-cardiac puncture. After recovery from anaesthesia, the animals were released at their capture sites. The blood samples from the animals were centrifuged (3000 x g, 15 min), and the sera obtained were aliquoted into 1.5 ml tubes and stored at $-20\,^{\circ}$ C.

2.3. Modified agglutination test and statistical analysis

The sera obtained from wild mammals were examined by the modified agglutination test – MAT (Desmonts and Remington, 1980). Animals with titres greater than or equal to 25 were considered positive (Gennari et al., 2015). Positive samples were diluted two-fold to obtain the final titre. In all reactions, previously known positive and negative mouse controls and an antigen control were used.

The differences in *T. gondii* seroprevalence between rodents and marsupials were analyzed by χ^2 association tests with $\alpha=5\%$ using Epi Info 6.0 software designed by the Centers for Disease Control and Prevention (Atlanta, Georgia).

2.4. Ethical statement

All the procedures followed the ethical standards of animal experimentation established by the Committee on Ethics and Studies and Research at the "Universidade Federal do Vale São Francisco" (CEDEP/Univasf; protocol numbers 0004/070813 and 0010/021014); and by the Brazilian Institute of Environment and Renewable Natural Resources (IBAMA; protocol numbers 36585-1 and 45764-1).

3. Results

A total of 75, 60 and 17 mammals were captured, respectively, in NPSC, Petrolina municipality and Lagoa Grande municipality. Among the marsupials, there were 25 females and 23 males; of those, 33 were adults and 15 were juveniles. Among the rodents, there were 41 females and sixty males; of those, 83 were adults and 18 were juveniles.

We found that 5.3% (4/75) and 3.3% (2/60) of small wild mammals were positive for IgG anti-*T. gondii* antibodies in the NPSC and Petrolina. All mammals from Lagoa Grande (0/17) tested negative on the MAT. The overall seroprevalence was 3.9% (6/152) (Table 1).

The prevalence of anti-T. gondii antibodies among animals of the

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