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### Short Communication

# Seroprevalence of anti-*Toxoplasma gondii* and anti-*Neospora caninum* antibodies in domestic mammals from two distinct regions in the semi-arid region of Northeastern Brazil



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#### ABSTRACT

The study was conducted to determine the seroprevalence of anti-*Toxoplasma gondii* and anti-*Neospora caninum* antibodies in goats, sheep, dogs and cats from two distinct regions in the semi-arid region of Northeastern Brazil: Serra das Confusões National Park (SCNP), a preserved area; and municipality of Petrolina, a non-preserved area. Overall, by the indirect fluorescent antibody test (IFAT), the prevalence for IgG anti-*T. gondii* antibodies in goats was 5.1% (19/376) and 2.4% (9/376); in sheep, it was 10.2% (34/332) and 14.2% (47/332); in dogs, it was 19.7% (25/127) and 3.9% (5/127) and, in cats, it was 25.7% (9/35) and 5.7% (5/35), for *T. gondii* and *N. caninum*, respectively. For *T. gondii* infection, the risk factors associated with the seroprevalence was gender (female) and study area (Petrolina) for goats and only the study area (Petrolina) for sheep. The circulation of *T. gondii* and *N. caninum* was observed in both areas, with significative risk factors demonstrated of a degraded environment. © 2016 Elsevier B.V. All rights reserved.

#### 1. Introduction

*Toxoplasma gondii* and *Neospora caninum* are intracellular parasites belonging to the phylum Apicomplexa. The two parasites have very similar structural characteristics, which rendered to a misdiagnosis of the parasite as *T. gondii* for many years (Dubey, 2003; Tenter et al., 2002).

*T. gondii* has a facultative heteroxenous life cycle that is optimal in that it can infect all warm-blooded animals. The protozoan is globally prevalent and highly relevant in veterinary medicine and public health for being a zoonosis, causing abortion and congenital diseases in intermediate hosts (Tenter et al., 2000).

Domestic and wild cats are the definitive hosts for *T. gondii*, the only ones that excrete the resistant oocysts in the feces (Dubey, 2009). Warm-blooded animals can be infected mainly by ingesting the oocysts contaminating the water and the environment in general, and by ingestion of cysts containing bradyzoites that develop in muscles and brain of infected hosts (Miller et al., 2009).

Neosporosis has emerged as a troubling disease that affects mainly cattle and dogs (Dubey, 2003). In recent years, it has been noted as a

\* Corresponding author. *E-mail address:* horta.mc@hotmail.com (M.C. Horta). major reproductive disease in cattle worldwide (Trees et al., 1999; Anderson et al., 2000; Dubey, 2003). Domestic dogs and some wild canids are the definitive hosts for *N. caninum* (Gondim et al., 2004; McAllister et al., 1998; King et al., 2010; Dubey et al., 2011a).

The possibility of transmission of *N. caninum* among wild and domestic animals has been widely discussed. The existence of a so-called wild cycle of neosporosis that may occur between canids and wild herbivores could influence the epidemiology of the disease in domestic livestock (Rosypal and Lindsay, 2005; Gondim et al., 2004).

The Caatinga biome is located in a semi-arid region of Brazil and has environmental conditions with sharp differences to the other Brazilian biomes (Arraes-Santos et al., 2015). It is characterized by rainfall irregularity from year to year, which result in severe droughts and an inhospitable environment for the survival of local fauna (Krol et al., 2001).

The present study aimed to evaluate the prevalence of anti-*T. gondii* and *N. caninum* antibodies in domestic mammals from two distinct areas within the Caatinga biome of Northeastern Brazil.

#### 2. Materials and methods

This study was conducted in two distinct areas within the Caatinga biome in the semi-arid region of Northeastern Brazil in the municipality of Petrolina, which is in the western part of the state of Pernambuco (9° 23′ 55″ S, 40° 30′ 03″ W) and in the National Park of Serra das Confusões (NPSC), which is in the southeastern part of the State of Piauí (9° 00′ 57″ S, 43° 26′ 51″ W) (Fig. 1). This biome, which is a mosaic of xerophilous, thorny bushes and forest that covers much of Northeastern Brazil and part of Minas Gerais, extends 735,000 km<sup>2</sup>. 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 municipality of Petrolina is located in the San Francisco Valley region, representing a medium-sized municipality within a degraded environment.

From January to October 2013 in NPSC and from August 2014 to April 2015 in Petrolina, blood was collected from domestic animals (goats, sheep, dogs and cats) from properties in villages at rural areas. Blood samples from sheep and goats were obtained by venipuncture of the jugular vein and from dogs and cats by venipuncture of the cephalic or jugular vein. After centrifugation (3000 g, 15 min), the obtained sera from each animal were aliquoted into 1.5 ml tubes and stored at -20 °C. All the procedures followed the ethical standards of animal experimentation established by the Committee on Ethics and Studies and Research at the Federal University of São Francisco Valley – CEDEP/Univasf (protocols numbers 0004/070813 and 0010/021014).

Antibodies against *N. caninum* and *T. gondii* from domestic animals were determined by indirect immunofluorescent antibody test (IFAT) according to Dubey et al. (1988) and Camargo (1974), respectively. The sera were tested at a start dilution and cutoff of 1:50 for *N. caninum* in all species; and at start dilutions and cutoff points of 1:16 in dogs and cats and1:64 in sheep and goats for *T. gondii*. In all reactions, positive and negative control sera were used.

An epidemiological questionnaire was conducted with the owners of domiciled animals concerning the following issues: age, breed, gender and management breed. We used the information obtained from the questionnaires in the analysis of risk factors.

The analysis of risk factors was performed in two steps: univariate and multivariate analysis. Univariate analysis was performed using the Chi-square test or Fisher's exact test, and those variables that presented  $P \le 0.20$  were used for multiple logistic regression. The multivariate analysis was then performed using the stepwise forward method

#### Table 1

Prevalence of *T. gondii* and *N. caninum* (IFAT) antibodies in goats from the Brazilian semiarid region.

Variables	Category	Number of goats	T. gondii		N. caninum	
			No. of positive (%)	P-value	No. of positive (%)	P-value
Region	Petrolina	174	12 (6.9)		5 (2.9)	
	NPSC <sup>a</sup>	202	7 (3.5)	$0.200^{*}$	4(2)	0.738
Goat breed	Pure breed	59	1 (1.7)		0(0)	
	Mixed breed	317	18 (5.7)	0.331	9 (2.8)	0.365
Age	6 months	155	3 (1.9)		5 (3.2)	
	to 1 year					
	1 to 3 years	155	12 (7.7)		2 (1.3)	
	>3 years	66	4 (6.1)	0.060*	2 (3)	0.501
Gender	Males	99	1(1)		2 (2)	
	Females	277	18 (6.5)	0.061*	7 (2.5)	1.000
Management breed	Extensive	69	4 (5.8)		1 (1.4)	
	Semi-intensive	248	14 (5.6)		8 (3.2)	
	Intensive	59	1 (1.7)	0.439	0 (0)	0.294

Variables selected for the multivariate analysis (P < 0.20).</li>

<sup>a</sup> National Park of Serra das Confusões.

(Hosmer and Lemeshow, 2000). The significance level in multivariate analysis was 5%. The adjustment of the final model was checked using the Hosmer and Lemeshow test, and  $P \ge 0.05$  was taken to indicate a satisfactory fit. The tests were performed using the SPSS software for Windows, version 20.0.

#### 3. Results

The prevalence of anti *T. gondii* and anti-*N. caninum* antibodies in goats, sheep, dogs and cats can be seen, respectively, in Tables 1, 2, 3 and 4.

The variables age, gender and region were selected ( $P \le 0.20$ ) for multivariate analysis (Table 1) of *T. gondii* in goats. The following variables were identified as risk factors by logistic regression



Fig. 1. Location of the two Brazilian states in which animals were sampled for the present study.

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