

**SEROPREVALENCE AND FACTORS ASSOCIATED OF  
*Toxoplasma gondii* INFECTION IN GOATS FROM THE  
PARÁ AND MARANHÃO STATES, BRAZIL**

*(Soroprevalência e fatores associados à infecção por Toxoplasma gondii  
em caprinos dos estados do Pará e do Maranhão, Brasil)*

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**RESUMO**

A epidemiologia da toxoplasmose em caprinos na região amazônica e pré-amazônica tem sido pouco estudada, havendo necessidade de mais estudos para evitar perdas na produção de pequenos ruminantes (queda de peso, baixo rendimento de carcaças e perdas reprodutivas face aos casos de abortamento), principalmente no âmbito familiar, pois a caprinocultura é a principal renda dos pequenos produtores. Soro de 412 caprinos nos estados do Pará e Maranhão foram submetidos a reação de imunofluorescência indireta para anticorpos contra *Toxoplasma gondii*. Do total de amostras testadas, 97 (23,5%) dos soros foram positivos para *T. gondii*. Não foi observada diferença estatística entre a soroprevalência no Estado do Pará e Maranhão. A idade dos animais apresentou uma relação direta com a soroprevalência de *Toxoplasma*. No Pará, propriedades onde gatos se alimentavam de restos de placenta dos caprinos e acessavam ao aprisco tinham 2,5 vezes mais chances de serem soropositivos para *T. gondii*. No Maranhão, propriedades com mais de seis gatos e caprinos com idade superior a 24 meses tinham 4,5 e 2,0 vezes, respectivamente, mais chances de serem soropositivo para *T. gondii*. Nossos resultados demonstram que o manejo sanitário dos animais deve levar em conta a quantidade e acesso de gatos aos caprinos, e impedir o acesso e alimentação de gatos com placenta de cabras. Animais com idade superior a 24 meses também foram considerados um importante fator de risco associado à infecção pelo *T. gondii* em caprinos.

**Palavras-chave:** *Toxoplasma gondii*, caprinos, reação de imunofluorescência indireta.

**ABSTRACT**

The epidemiology of toxoplasmosis in goats in the Amazon and pre-Amazon regions has been scarcely investigated, indicating that further studies are necessary to avoid losses in the production of small ruminants (due to factors such as: weight loss, low carcass yield and reproductive losses in the face of abortion cases), especially in the family context, as goat farming is the major income of small producers. Serum from 412 goats in Pará and Maranhão states were subjected to the *Toxoplasma* immunofluorescence antibody test for antibodies against *Toxoplasma gondii*. Of the total sample tested, 97 (23.5%) sera were positive for *T. gondii*. No statistical difference was found between the seroprevalence in Pará and Maranhão. The age of the animals showed a direct relationship with the prevalence of *Toxoplasma* infection. In Pará, farms where cats are fed placental remains and have access

to the stable are 2.5 times more likely to be seropositive to *T. gondii*. In Maranhão, farms with more than six cats and those with goats older than 24 months of age are 4.5 and 2.0 times, respectively, more likely to be seropositive for *T. gondii*. Our results demonstrate that the sanitary handling of animals must consider the amount and access of cats to goats, and how to properly prevent the access and feeding of cats with placenta of goats. Animals older than 24 months were also considered an important risk factor associated with infection by *T. gondii* in goats.

**Key words:** *Toxoplasma gondii*, goat, immunofluorescence antibody test.

## INTRODUCTION

Toxoplasmosis is one of the most common parasitic zoonoses in the world. *Toxoplasma gondii* has a facultative heteroxenous life cycle and can infect all warm-blooded animals (mammals and birds). The parasite is mainly transmitted by oral ingestion of oocysts from the environment, horizontally by oral ingestion of tissue cysts of intermediate hosts, or vertically by transplacental transmission of tachyzoites (TENTER *et al.*, 2000; DUBEY, 2009).

Infection caused by *T. gondii* in pregnant goats may cause early embryonic death and resorption, fetal death and mummification, abortion, stillbirth and neonatal death, resulting in reproductive and economical losses. However, the infection has public health implications since consumption of infected meat can facilitate zoonotic transmission (JITTAPALONG *et al.*, 2005).

Brazil has 9.5 million head of goats, around 1.1% of the total number of goats in the world (FAO, 2008), and 90.6% of this total are concentrated in the semi arid, and pre Amazon regions where this species is an important source of meat and milk for humans. In the Amazon region the goat herd grew 19.4% between 2004 and 2009 and may be an alternative to animal production in this region with low environmental impact (BRAZIL, 2017).

Worldwide estimates of the sero-prevalence of goat toxoplasmosis are highly variable depending on the serologic test used, the region under study, and the age of the animals (TENTER *et al.*, 2000). In Brazil, serological surveys show infection rates ranging between 10.0% (AMARAL *et al.*, 1978) and 92.1% (CHIARI *et al.*, 1987). In Semi-Arid studies were performed in almost all states (RAGOZO *et al.*, 2009) but rare are the studies in the Pre Amazon (Maranhão State) and Amazon (Pará state) (BRANDÃO *et al.*, 2009). Due to the insufficient data regarding goat culture in the Amazon and Pre Amazon of Brazil, and also because it is a serious disease with impacts on public health and reproduction, the purpose of this study was to estimate the prevalence of *T. gondii* in goats of Pará (Amazon of Brazil) and Maranhão (Pre Amazon of Brazil) and to identify associated factors.

## MATERIAL AND METHODS

### Study area

All procedures were performed with approval by the Animal Ethics Committee – UFMA (process nº 23115.005441/2017-62). The universe of samples comprehend the properties listed by Associação dos Criadores de Caprinos e Ovinos do Pará (ACCOPA),

Agência Estadual de Defesa Agropecuária do Pará (ADEPARÁ) and Agência Estadual de Defesa Agropecuária do Maranhão (AGED/MA). These sources of information were used to identify the main areas of production and greater representation of goat herds within the communities that constitute the region under study. The area selected for sample collection included four farms in the state of Pará in Brazilian Amazon and ten farms in Chapadinha city, state of Maranhão, in Brazilian Pré Amazon (Fig. 01). In Pará, the farms have mean of 100 animals, mixed breed, and maintained in cultivated pasture without supplementation. In Maranhão, the farms have mean of 80 animals, mixed breed, and 70% are maintained in native pasture without supplementation.



**Figure 01:** Geographic location of studied municipalities.

### **Animals**

The sample size was calculated using formula of Centro Panamericano de Zoonosis (1973), with an expected prevalence of 30.6% according Araújo-Neto *et al.* (2008), with a confidence interval of 95% and an error margin of 15%, giving a total of 387.2. However, an extra 6% samples was collected. 10% of each animal category (kids goats, adult males, and females empty or pregnant) were randomly sampled in each farm, being collected at least 10 animals per farm.

A total of 412 mixed breed goats were analyzed. Blood was collected from the jugular vein and the serum was separated after blood cell sedimentation and stored at -20 °C until use. At the time of blood collection the owner of each farm was interviewed using a pre-coded questionnaire especially designed for this study. Information on the farm and herds was collected including: (1) education level of the farmers; (2) characterization of each farm (water supply, type of water facility, feeding troughs, land use (extensive/intensive), abortion, and presence and types of barn; (3) presence of cats (number of domestic cats, access of cats to the water supplies, feeding the cats with placental remains, and control of rodents); and (4) individual animal data (age and sex of each animal sampled).

### **Indirect Fluorescence Antibody Test (IFAT)**

The indirect immunofluorescence antibody test (IFAT) was prepared and used as described previously (CHIARI *et al.*, 1987). Titers of 1:64 or above were considered to be positive as previously recommended (FIGUEIREDO *et al.*, 2001).

### Statistical Analysis

The results were analyzed by 2 x K contingency tables of exposure variables. The outcome variable was seropositivity to *T. gondii* and the independent variables were as follows: individual animal data (age and sex); presence of cats (number of domestic cats, access of cats to the water supplies, feeding the cats with placental remains); control rodents; cat birth control; and access of the cats to the stable. Odds Ratios (ORs) and *P*-values were calculated separately for each variable using the BioEstat software (AYRES *et al.*, 2003), and logistic regression was performed with a *P*-value <0.20 for variables in univariate analysis using Minitab 14 for Windows.

## RESULTS AND DISCUSSION

A total of 97 (23.5%) out of the 412 goats were found to be seropositive for *T. gondii*. Prevalence rates in Pará and Maranhão were 21.1% (34/161) and 25.1% (63/251), respectively which are lower than those found in Iran, which the overall prevalence rate was estimated to be 27% in goats (SHARIF *et al.*, 2015). Within Brazil, our results are higher than those observed in Paraíba (Northeastern), 11.1% (CORREIA *et al.*, 2015) and lower than those noticed in Pernambuco, 25,86% (LUCIO *et al.*, 2016). No statistical difference was found between the seroprevalence in Pará and Maranhão (*p*=0.41). In all farms animals were found to be seropositive for *T. gondii*.

Goats who were handled in semi-extensive systems (State of Pará) presented 6.5% positivity. Just as in the State of Maranhão, the extensive system stood out and the positive results obtained were 25.8%. The meaningful association between the semi-intensive handling of livestock and the presence of *T. gondii* in goats shows the increase in contact risk for the agent of transmission. Tenter *et al.* (2015) showed that the prevalence of toxoplasmosis is related to the system adopted for handling the animals. The semi-intensive productive systems employed in the farming of sheep and goats may present low healthiness conditions allowing the dissemination of *T. gondii* oocysts.

The variables of sex, presence of cats, number of cats, access of the cats to water supplies of the goat, feeding of cats with placental remains, access of the cats to goats, access of the cats to other farms, access of the cats to the stable, cat birth control and rodent control did not constitute risk factors related to the seroprevalence of *T. gondii* in both regions. Sporulated oocysts of *T. gondii* contained in the environment are a potential source of infection and are very resistant to environmental conditions (TENTER *et al.*, 2000). Hamada *et al.* (2019) reported that toxoplasma seropositivity in goats and sheep increases when the presence of cats is frequent in the herds feeding places.

Here we show that goats that live in Pará farms where cats have access to the stable (and likely can contaminate it) are 2.5 times more likely to be seropositive to *T. gondii* than other animals. Similarly, we showed that farms in Maranhão state with six or more cats are 4.5 times more likely to have goats seropositive for *T. gondii*.

Statistical differences were observed in the relationship between anti-*T. gondii* antibodies and the age of the goats, indicating that animals over 24 months old have undergone more risk of infection when compared to animals at earlier ages. Some studies

have demonstrated that animals at older ages present higher positive rates due to the fact that they stay exposed to *T. gondii* oocysts in the environment for a longer time. The presence of such animals, which are bearers of infection, among others in herds where the prevalence of the infection is high may be considered an importance risk factor to *T. gondii* infection (SELLA *et al.*, 1994; FIGLIULO *et al.*, 2004).

**Table 01:** Frequency rate and factors associated of *Toxoplasma gondii* infections in goats from the Pará, Amazon region and Maranhão, Pre-Amazon region states.

	<b>Variable</b>	<b>% Prevalence (+ / n)</b>	<b>Adjusted OR (95% CI)</b>	<b>P-value</b>
<b>Age</b>	<12 months	16,2 (20/123)	Ref.	Ref.
	12–24 months	23,3 (31/133)	0.85 (0.83-2.92)	0,20
	>24 months	29,4 (46/156)	0.73 (1,19-3,88)	0,01
<b>Sex</b>	Male	18,9 (24/127)	Ref.	Ref.
	Female	25,6 (73/285)	1.47 (0,88-2,48)	0,17
<b>Cats<sup>a</sup></b>	Absence	17,2 (5/29)	Ref.	Ref.
	Presence	24,0 (92/383)	1.51 (0,56-4,09)	0,54
<b>Nº of Cats<sup>b</sup></b>	Absence	17,2 (5/29)	Ref.	Ref.
	1–2 cats	19,5 (28/143)	1,16(0,40-3,33)	0,97
	3–5 cats	18,7 (20/107)	1,10 (0,37-3,24)	0,92
	≥6 cats	33,1 (44/133)	2,37 (0,84-2,48)	0,14
<b>Access to Stalls</b>	No	21,5 (67/311)	Ref.	Ref.
	Yes	30,0 (30/101)	1,53 (0,92-2,55)	0,12
<b>Rodent Control</b>	Yes	30,0 (30/101)	Ref.	Ref.
	No	21,5 (67/311)	0,64 (0,39-1,07)	0,12
<b>Access to Water</b>	No	24,9 (93/373)	Ref.	Ref.
	Yes	2,6 (1/39)	0,07 (0,01-0,58)	0,01
<b>Feed on placentar remnants</b>	No	21,5 (67/311)	Ref.	Ref.
	Yes	30,0 (30/101)	1,53 (0,92-2,55)	0,12
<b>Birth Control</b>	Yes	29,3 (22/75)	Ref.	Ref.
	No	21,4 (72/337)	0,65 (0,37-1,14)	0,18
<b>Access to Neighboring Property</b>	No	23,0 (79/344)	Ref.	Ref.
	Yes	26,4 (18/68)	1,20 (0,66-2,18)	0,64
<b>Access to Cat</b>	No	24,1 (63/261)	Ref.	Ref.
	Yes	22,5 (34/151)	0,91 (0,56-1,46)	0,80
<b>Total Frequency</b>		23,50 (97/412)		

+: Number of positive animals; n: Number of samples per variables; OR: Odds Ratios; P: Probability; 95% CI: 95% Confidence Interval; Ref: Variable used as reference value.

In Pará, farms where cats are fed the placental remains and have access to the stable are 2.5 times more likely to be seropositive to *T. gondii*. The others variables studied did not constitute risk factors. In Maranhão, farms with more than six cats and those with goats older than 24 months of age are 4.5 and 2.0 times, respectively, more likely to be seropositive to *T. gondii* than other variables (Tab. 01, above). Organotropism of *T. gondii* and the number of tissue cysts produced in a certain organ vary with the intermediate host species. *T. gondii* tissue cysts are most frequently observed in various tissues of infected pigs, sheep and goats

(TENTER, 2009). Possible routes of transmission are considered include ingestion of bovine fetuses and cattle (VAN RENSBURG e SILKSTONE, 1984; SPENCER *et al.*, 2003).

## CONCLUSIONS

Our results demonstrate that the management of the animals' health has to take into account the number of cats (in the area) and their access to the goats. The associated factors were different in each region studied. Therefore, more epidemiological studies are required to identify the sources of infection, the impact on animal breeding in the region, and the risk of transmission to humans

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