

Prevalence and geographical distribution of *Toxoplasma gondii* in dogs in the urban area of Botucatu, SP, Brazil

Prevalência e distribuição geográfica do Toxoplasma gondii em cães na área urbana de Botucatu, SP, Brasil

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Abstract

Toxoplasmosis is a worldwide zoonosis caused by *Toxoplasma gondii*, and can infect a wide variety of animals including humans. Domestic animals can be an important sentinel population for infection in the community. Occurrence of *T. gondii* infection was assessed in dogs in the urban area of Botucatu city, SP, Brazil. In the sample, 10% rate for error estimate, 95% confidence interval, and 5% significance level were established. Serum samples were collected from dogs during a rabies vaccination campaign, and were processed using modified agglutination test (MAT). Blood samples were collected from 670 dogs, with homogeneous distribution in five regions in the urban area, representing 3.74% of 17,910 animals vaccinated. In this sample, 17.3% (116/670) dogs (68 – 58.6% female and 48 – 41.4% male) were positive for *T. gondii* infection ($p<0.03$). Regarding age of the infected dogs 4.6% (4/88) were younger and 95.4% (84/88) were older than one year ($p<0.01$); the age of 28 positive animals were undetermined. The serum titers of anti-*T. gondii* antibodies were: 16 (69.8%; 81/116), 64 (13.8%; 16/116), 256 (15.5%; 18/116), and 1024 (0.9%; 1/116). Prevalence was distributed among the North 14.2% (19/134), South 18.0% (31/172), East 15.7% (19/121), West 21.6% (27/125), and Central 16.9% (20/118) regions of the municipality ($p=0.5$). In all these regions, females and dogs aged more than one year showed a higher occurrence of *T. gondii* infection ($p<0.05$).

Keywords: Dogs. *Toxoplasma gondii*. Spatial distribution.

Resumo

Toxoplasmose é uma zoonose de distribuição mundial causada pelo *Toxoplasma gondii*, que pode infectar uma grande variedade de animais, inclusive seres humanos. Animais domésticos podem ser sentinelas importantes para infecções na comunidade. Ocorrência de infecção pelo *T. gondii* foi avaliada em cães na área urbana de Botucatu (SP, Brasil). Para amostragem, considerou-se uma taxa de erro na estimativa de 10%, um intervalo de confiança de 95% e um nível de significância de 5%. As amostras de sangue dos cães foram coletadas durante uma campanha de vacinação antirrábica e processadas usando-se o teste de aglutinação modificado (MAT). Foram coletadas 670 amostras de sangue, com distribuição homogênea em cinco regiões da área urbana, representando 3.74% dos 17.910 cães vacinados. Dessa amostra, 17,3% (116/670) dos cães, sendo 58,6% (68/116) fêmeas e 41,4% (48/116) machos, foram positivos para infecção pelo *T. gondii* ($p<0,03$). Entre os cães infectados 4,6%; (4/88) deles tinham idade menor e 95,4% (84/88) maior que um ano ($p<0,01$); em 28 animais positivos a idade não era conhecida. Os títulos séricos de anticorpos anti-*T. gondii* estavam distribuídos entre 16 (69,8%; 81/116), 64 (13,8%; 16/116), 256 (15,5%; 18/116) e 1024 (0,9%; 1/116). Os animais positivos estavam distribuídos nas regiões Norte 14,2% (19/134), Sul 18,0% (31/172), Leste 15,7% (19/121), Oeste 21,6% (27/125) e Centro 16,9% (20/118) do município ($p=0,5$). Em todas essas regiões, fêmeas e animais com mais de um ano de idade tiveram maior taxa de positividade para a infecção ($p<0,05$).

Palavras-chave: Cães. *Toxoplasma gondii*. Distribuição espacial.

Toxoplasmosis is a zoonotic disease of worldwide distribution, which can infect a variety of species and man. The first reports on the presence of *Toxoplasma gondii* in dogs describe its occurrence in Italy (Mello, 1910) and in Brazil (Carini, 1911; VIDOTTO)¹. Dogs are highly susceptible to infection². Carnivores such as dogs are infected by ingesting meat from animals

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containing cysts of the agent. Herbivores are infected by ingesting oocysts that are present in pastures². Investigation on animal toxoplasmosis is essential due to its transmission to humans and pathogenicity in pets and production animals³. From the viewpoint of public health, infection of dog population means that the area involved is an ecological niche for this coccidian therefore being a risk to the human population⁴. Although dogs are not definitive hosts of this protozoan, they contribute to the mechanical dissemination of this protozoonosis⁵. Similarly to what occurs in humans, infection in dogs is most often asymptomatic and can cause reactivation of chronic cases when there is immunosuppression⁶. The direct agglutination test has been utilized to show anti-*T. gondii* agglutinin in various species of domestic and wild animals⁷. Therefore, the objectives of present study were to analyze the prevalence of toxoplasmosis in dogs in Botucatu city (SP, Brazil) and geographical distribution of the infection as a contribution to the study of epidemiological chain of toxoplasmosis in this region.

Botucatu city (1,483 km²) is in the Center-South region of the state of São Paulo, and its dog population was estimated to be 29,923 animals (2002)⁸. A total of 670 samples of blood were collected from male and female dogs with different ages during animal rabies vaccination (3.74% of 17,910 vaccinated dogs) in 2004. The samples were collected in 20 of the 47 vaccination posts, which were homogeneously distributed in five regions of the urban area. Posts that included blood collection were geographically randomized. For systematic collection of samples, every third animal brought for vaccination was selected for blood collection. The sample size was established, accepting a 10% rate of the estimate error and 95% confidence interval. The direct agglutination test (MAT) was used for detection of anti-*T. gondii* antibody⁹, and samples with titers ≥ 16 were considered positive⁷. Information was stored in a database and analyzed

with the EpiInfo 3.4.3 software, using the odds ratio (OR), 95% confidence interval (CI95%), and $P \leq 0.05$.

In the sampled population, 49.9% (334/670) males and 48.2% (323/670) females was observed and in 1.9% (13/670) dogs the gender were undetermined. Regarding age range, 9.7% (65/670) of these animals were younger and 64.9% (435/670) were older than one year and in 25.4% (170/670) the age of the dogs was not determined. Geographical distribution of the dogs was 20.0% (134/670), 25.4% (172/670), 18.4% (121/670), and 18.6% (125/670) in the North, South, East, and West of the municipality, respectively, and 17.6% (118/670) in the Central area of Botucatu.

A total of 116 samples positive for *T. gondii* were detected, with 17.3% prevalence (95%CI: 14.6-20.5). Titers 16, 64, and 256 were found in 69.8, 13.8, and 15.5% (95% CI: 60.6-78.0, 8.1-21.4, and 9.5-23.4) of the seropositive samples, respectively. The highest value for titer (1024) represented 0.9% of the samples (95% CI: 0-4.7). Germano, Erbolato, and Ishizuka⁴ used the indirect immunofluorescence antibody test (IFAT) in a serological study conducted in Campinas (SP, Brazil) and found 91% of positive results. Garcia et al.³ also used IFAT to analyze 189 samples of dog serum in Jaguapitã (PR, Brazil), of which 84.1% were reactive. Sixteen and 64 were the most frequent titers (31.4 and 38.4 % of samples, respectively). The highest titer (4.096) was found in 1.9% of the samples. Da Silva, Cutolo, and Langoni⁷ used MAT in Botucatu (SP, Brazil) and observed 19.0% of positive results in samples with varying titers in the range 16-256. The same test was also used by Cañón-Franco et al.¹⁰, who found 85% of positive dogs in Monte Negro (RO, Brazil), and Da Silva et al.¹¹, who found 25.4% (52/205) in Ubatuba (SP, Brazil). Barbosa et al.¹² used IFAT for analysis of anti-*T. gondii* antibodies in sera (225 samples) of stray dogs and found 63.5% of positive results; titers ranged from 16 to 16.384, and titer 64 was found in 44.7% of samples. The studies mentioned above reported *T. gondii* infection in

Table 1 - Results of the modified agglutination test (MAT) for anti-*T. gondii* antibody in dog serum, by gender and age group (Botucatu, SP, Brazil, 2012)

Variables	Positive (%)	Negative (%)	OR	95% CI	<i>p</i> values
GENDER (n=657)					
Female	68 (58.6)	255 (47.2)	1.6	1.05-2.38	<0.03
Male	48 (41.4)	285 (52.8)			
AGE (months) (n=500)					
> 12	84 (95.4)	351 (85.2)	3.6	1.29-10.34	<0.01
≤ 12	4 (4.6)	61 (14.8)			

OR: odds ratio; 95% CI: 95% confidence interval

dogs and showed variable results, regardless of the serological technique. However, most of them showed titers and occurrence higher than those found in the present study.

Regarding gender, 41.4% (48/116) of males and 58.6% (68/116) of females were seropositive and thus the probability of seropositive animals being females was 1.6 times higher than that in seronegative animals. In 28 samples the age of the dogs was undetermined and in the others the probability of seropositive dogs being older than one year was 3.6 times greater than in seronegative dogs. Table 1 indicates that age group ($p < 0.01$) and gender ($p < 0.03$) had a significant association with *T. gondii* occurrence. Thus, our findings regarding age are similar to those obtained by Jackson, Hutchison, and Siim¹³ and Garcia et al.³, who showed a lower seropositivity in dogs aged less

than six and eight months, respectively. As noted by Barbosa et al.¹², a significant association was observed in relation to age group.

Canine toxoplasmosis was distributed throughout the municipality. Prevalences in the North 14.2% (19/134), South 18.0% (31/172), East 15.7% (19/121), West 21.6% (27/125) regions and in the city center 16.9% (20/118) showed no statistical difference ($p = 0.5$).

Therefore, we conclude that prevalence of *T. gondii* infection in dogs in the urban area of Botucatu city was low and its distribution was homogeneous. Our data point to the importance of serological surveys since they are an important tool in the surveillance, control of infections, such as toxoplasmosis, and assessment of risk to humans. Furthermore, they indicate the importance of dogs as sentinel animals.

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