

Prevalence and Risk Factors of Toxoplasmosis among Pregnant Women in Fortaleza, Northeastern Brazil

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Abstract. We determined the seroprevalence of *Toxoplasma gondii* and associated risk factors among 963 pregnant women attending an obstetric hospital in Fortaleza, Brazil. Seroprevalences of IgG and IgM against *T. gondii* were 68.6% (95% confidence interval [CI] = 65.6–71.6%) and 0.5% (95% CI = 0.06–1.0%), respectively. Seroprevalence of IgG was high in women less than 25 years of age (91.7%) and in low-income women (odds Ratio [OR] = 1.40, 95% CI = 1.02–1.90). Multivariate regression analysis showed that consumption of homemade water ice (adjusted OR = 1.49, 95% CI = 1.09–2.04), vegetables washed with untreated water (adjusted OR = 1.43, 95% CI = 1.05–1.94), consumption of chicken (adjusted OR = 1.49, 95% CI = 1.12–2.0), and dog ownership (adjusted OR = 1.46, 95% CI = 1.07–1.98) were factors associated with IgG seropositivity. Young women in northeastern Brazil living under poor socioeconomic conditions are at highest risk for acquiring infection with *T. gondii*. Oocyst contamination of water and soil must be addressed in future prevention strategies.

INTRODUCTION

Approximately one-third of the world's population is infected with the protozoan parasite *Toxoplasma gondii*.¹ Humans are commonly infected by oral ingestion of water, food, or soil contaminated with oocysts, or by consumption of raw and undercooked meat contaminated with cysts of *T. gondii*. Placental transmission to the fetus may occur in women acquiring primary maternal infection during pregnancy. The rate of transplacental transmission and severity of disease in the fetus are inversely correlated.^{2,3} Whereas early in pregnancy, the rate of transmission is low, clinical manifestations are severe and may result in death of the fetus and spontaneous abortion. Late maternal infection is associated with a high rate of transmission, but clinical signs in the newborn are mild or absent upon first examination. However, serious sequels such as neurologic impairment and chorioretinitis can manifest in the second or third decade of life.^{4–6} Incidence of congenital toxoplasmosis ranges from 1 to 10/10,000 live births.⁶ In Brazil, congenital toxoplasmosis is observed in approximately 3.3–8/10,000 newborns.^{7–12}

The seroprevalence of infection with *T. gondii* is influenced by cultural, hygienic, and nutritional habits, and by climate and environmental conditions.^{1,5} Low rates of seropositivity in pregnant women and women in childbearing age have been reported in northern Europe and the United States.^{13–15} Conversely, most studies from central and southern America report high seroprevalence.³ A recent study in pregnant women in Colombia showed a seroprevalence of 49%.¹⁶ The seropositivity in Brazil ranges from 51% to 71% for pregnant women and women of childbearing age.^{17–23}

Detailed knowledge on the prevalence and risk factors of infection with *T. gondii* are required to design appropriate prevention measures against infection during pregnancy and congenital transmission. In the present study, we therefore

investigated seroprevalence of infection with *T. gondii* and associated risk factors in a large number of pregnant women attending a public tertiary care obstetric hospital in northeastern Brazil.

MATERIALS AND METHODS

Study design and population. This cross-sectional study was conducted in Fortaleza, the fourth largest city in Brazil. This city has a population of 2.4 million, is the capital of the state of Ceará, and is located in the dry northeastern region of the country. The climate is dominated by more than 90% half-arid regions with slight rainfalls. Fortaleza is located on the coast and had a hot and dry climate; seasonable differences are small. The average temperature is 26.5°C (range = 23°C–28°C). The average annual precipitation is 1,600 mm.

All pregnant women admitted for delivery to the Maternidade Escola Assis Chateaubriand (MEAC) of the Medical Faculty of the Federal University of Ceará from February 22 through May 4, 2005 participated on the study. The MEAC is a reference center for gynecology and obstetrics. It has 235 beds, a neonatal and maternal intensive care unit, and units for maternal–fetal medicine, obstetrics, and high-risk pregnancies. The number of births per year at this facility is approximately 4,800. Medical care is provided free; the hospital therefore serves mainly patients living in poor communities in the urban, suburban, and rural communities of Fortaleza.

Inclusion criteria for the study were attendance for delivery at the MEAC, residency in Fortaleza or in the metropolitan region surrounding the city, and written consent of the patient or of a legal guardian in women less than 18 years of age.

To determine the seroprevalence of infection with *T. gondii*, venous blood was obtained from study participants at the MEAC. Blood was not obtained from women who had IgG against *T. gondii* (5% of the pregnant women). This value was determined during prenatal care in the ambulatory setting.

Questionnaires. A pretested, structured questionnaire was used to assess demographic, socioeconomic, and behavioral variables. Women were interviewed by one of three investigators

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(S.S., N.B., or A.W.). Questions focused on possible risk factors for infection, including the presence or ownership of animals, eating habits, soil contact, and drinking water sources. The questionnaire was adapted from two studies conducted in northeastern and southern Brazil.^{21,24}

Serologic analysis for *Toxoplasma*. Serum samples were tested for IgG and IgM against *T. gondii* by using a micro-particle enzyme immunoassay (AxSym Toxo Assay; Abbott Laboratories, São Paulo, Brazil). Serologic tests were performed at the central laboratory of the university hospital according to the manufacturer's instructions. Women with positive IgG titers but negative IgM titers were considered latently infected. Women with positive IgG and IgM titers were considered to have a possible recent infection. In this case, a serum sample from the newborn was obtained either from the umbilical cord or from a peripheral vein and tested for IgM.

Statistical analysis. Data were entered into a database using Epi-Info version 6.04 software (Centers for Disease Control and Prevention, Atlanta, GA) and checked for entry errors. With an estimated prevalence of 60–70% and a 95% confidence interval (CI), a sample size of at least 814 women was estimated before data collection. Bivariate analysis with calculation of the odds ratio and respective 95% CIs was performed with STATA version 7 software (Stata Corp., College Station, TX). Because of the low number of IgM-positive women, bivariate analysis was conducted for IgG serostatus. Fisher's exact test was applied to determine significance of difference of relative frequencies. Multivariate logistic regression with backward elimination was used to calculate adjusted odds ratios for the independent association between infection with *T. gondii* (presence of IgG) and potential risk factors. Exclusion criteria from data analysis were absence of a questionnaire, an incomplete questionnaire, lack of a serum samples, or an insufficient quantity of serum. Data from women with known positive IgG *T. gondii* antibody status on the basis of prenatal care were included in analysis.

Ethics. Ethical approval for the study was obtained from the Ethical Review Board of the Medical Faculty, Federal University of Ceará. All participants were informed about the study, and informed written consent was obtained from all study participants, or in case of minors, from their guardians.

RESULTS

A total of 1,000 pregnant women were enrolled, and data of 963 women were analyzed. The datasets of 37 women were excluded because of absence of a questionnaire, an incomplete questionnaire, lack of serum samples, or insufficient quantity of serum. Mean age of participants was 25.2 years (range = 12–44 years). Of the 963 women, 661 (68.6%, 95% CI = 65.6–71.6%) had IgG against *T. gondii* and only 5 (0.5%, 95% CI = 0.06–1.0%) had IgM against *T. gondii*. Age-dependent IgG seroprevalences are shown in Table 1. In the youngest age group (12–15 years), 91.7% of the patients had IgG against *T. gondii*. In the oldest age group (31–44 years), 77% of the patients had IgG against *T. gondii*.

Bivariate analysis showed a significant association between an age >25 years and IgG seropositivity (Table 2). Low monthly income and living in poor housing conditions, including streets made of sand and houses with sand floors, were also associated with IgG seropositivity. In contrast, living in Fortaleza or in the metropolitan region, illiteracy, or the lack of employment were not significantly associated with IgG seropositivity.

TABLE 1
Seroprevalence of *Toxoplasma gondii* among pregnant women, stratified by age group, in Fortaleza, Brazil

Age group, years	No. of IgG-positive women/total	Seroprevalence, % (95% confidence interval)
12–15	11/12	91.7 (61.5–99.8)
16–18	38/104	63.5 (53.4–72.7)
19–25	158/438	63.9 (59.2–68.4)
26–30	57/132	69.8 (62.8–76.3)
31–44	44/147	77.0 (70.3–82.7)
Total	661/963	68.6 (65.6–71.6)

Bivariate analysis of animal and soil contacts showed that ownership of dogs, but not ownership of cats, contact with cats, or contact with soil while working outside, were significant risk factors (Table 3). In contrast, frequent consumption of chicken and the consumption of homemade water ice were significantly associated with seropositivity.

Multivariate logistic regression showed that earning less than one minimum wage (U.S. \$120) was significantly associated with IgG seropositivity (Table 4). In addition, the consumption of chicken more than twice a week and the consumption of vegetables washed with untreated water were significantly associated with IgG seropositivity. Interestingly, the consumption of homemade water ice (locally called *dindin*, which is usually prepared with water) was also significantly associated with *T. gondii* infection.

The five patients positive for IgM against *T. gondii* were < 25 years of age, and consumed meat such as chicken more than two times a week, vegetables washed with untreated water, and homemade water ice. Three of five newborns whose mothers were IgM seropositive did not have IgM against *T. gondii*. In one case, the mother did not permit a blood sample to be obtained from the newborn. In a second case, another newborn was lost to follow-up. No follow-up samples were available for the first year of life.

DISCUSSION

Infection with *T. gondii* has a high prevalence in South America. Whereas acute infection in most persons is asymptomatic or mild, infection during pregnancy may cause transmission to the fetus with severe signs and symptoms.⁶ The infection is commonly acquired by the oral route and thus represents a preventable infection. However, to develop appropriate prevention programs, detailed knowledge on prevalence and risk factors is needed.

In the present study, we screened more than 950 pregnant women attending a tertiary care obstetric hospital in northeastern Brazil for antibodies against *T. gondii* and investigated potential risk factors by using a pretested questionnaire. Most interestingly, we observed a high IgG seroprevalence of 91.7% in young persons (12–15 years of age). In contrast, few pregnant women had IgM against *T. gondii*; all of these women were < 25 years of age. A high seroprevalence of infection with *T. gondii* in children (40%) and teenagers (60%) has been reported in Fortaleza and others parts of Brazil.^{18,25–27} The overall prevalence in pregnant women from our study was 69%, which was slightly higher than that observed in teenagers. The increase in prevalence with age is consistent with results of previous studies.^{18,19,23,28} Furthermore, similar results from other studies in northeastern Brazil have

TABLE 2

Bivariate analysis of socioeconomic factors associated with infection with *Toxoplasma gondii* in 963 pregnant women in Fortaleza, Brazil*

Risk factor	No.	No. (%) IgG positive	Odds ratio (95% confidence interval)	P
Socioeconomic factors				
Age, years				
≥ 25	380	279 (73.4)	1.52 (1.13–2.05)	0.004
< 25	554	357 (64.4)		
Education				
Illiterate	28	22 (78.6)	1.70 (0.66–5.18)	0.305
Literate	934	638 (68.3)		
Monthly household income, MW				
≤ 1	379	278 (73.4)	1.49 (1.11–2.01)	0.007
> 1	566	367 (64.8)		
Employment				
No	618	431 (69.7)	1.13 (0.84–1.52)	0.424
Yes	343	230 (67.1)		
Residence/location				
Metropolitan region of Fortaleza	77	52 (67.5)	0.93 (0.55–1.61)	0.798
Fortaleza	868	599 (69.0)		
Street				
Sand/cobblestone	359	262 (73.0)	1.39 (1.03–1.88)	0.026
Asphalt	603	398 (66.0)		
House				
Loam	20	16 (80.0)	1.85 (0.59–7.65)	0.336
Brick	943	645 (68.4)		
Floor of the house				
Sand/soil	34	29 (85.3)	2.73 (1.02–9.10)	0.037
Cement/ceramic	929	632 (68.0)		
Sanitary conditions				
Forrest/pothole	538	376 (69.9)	1.16 (0.87–1.54)	0.294
Sewage pipe	420	280 (66.7)		
Contact with soil†				
Yes	116	81 (69.8)	1.07 (0.69–1.68)	0.831
No	847	580 (68.5)		
Cat ownership				
Yes	246	179 (72.8)	1.31 (0.94–1.83)	0.112
No	716	481 (67.2)		
Dog ownership				
Yes	360	264 (73.3)	1.43 (1.06–1.92)	0.018
No	603	397 (65.8)		

*MW = minimum wage of 260 Reais = U.S. \$120.

† Contact with soil while working in the garden outside with plants.

indicated that seroprevalence has not changed in the past 10 years.^{17,18,21}

Recently, the prevalence of infection with *T. gondii* has been linked to the socioeconomic status in several studies.^{20,23,29} For example, 84% of the population of Rio de Janeiro, Brazil with low socioeconomic status was seropositive for *T. gondii*, compared with 62% with middle socioeconomic status and 23% with high socioeconomic status.²⁴ Similar findings were reported from Columbia.¹⁶ A higher seroprevalence has also been observed in public hospitals attended by persons with low incomes compared with private hospitals in Brazil.³⁰ Our study population was already biased towards lower socioeconomic status because we obtained data in a public university hospital that provides free care. Nevertheless, socioeconomic variables were found to be significantly associated with infection. In resource-poor communities, infection appears during childhood and early adulthood. These findings explain the high seroprevalence (> 90%) even in the youngest age groups in our patients and the low prevalence of IgM against *T. gondii* in the absence of congenital infections. In a similar study, Barbosa and others detected IgM against *T. gondii* in 1 of 190 pregnant women in Natal, which is also in northeastern Brazil.²³ Whereas the number of potentially acutely infected women who had IgM against *T. gondii* in our study was too low to perform statistical analyses, all women who were IgM

positive were 15–23 years of age and showed exposure variables similar to the risk factors for infection identified in the IgG-positive pregnant women.

Consumption of the homemade water ice (dindin) was one of the strongest risk factors for seropositivity against *T. gondii* (OR = 1.49). Dindin is a semi-commercial product produced in small home factories and is made with water, fruits, and sugar. The product is then transferred to small plastic bags and frozen. Interestingly, in a recent epidemiologic study in a rural region 40 km from Fortaleza, we also identified this homemade water ice as a significant risk factor for infection with *T. gondii* in pregnant women, which supports the assumption that there may be a causal relationship between consumption of dindin and infection with *T. gondii*.²¹ Contamination by oocysts of water used in household manufacturing is the most likely source of infection. The contamination of plastic bags rather than the water ice could also attribute to infection. Survival of cysts may occur in water that is not sufficiently frozen to kill oocysts. In this regard, investigation of an outbreak of toxoplasmosis in southern Brazil also identified consumption of ice prepared with contaminated drinking water as a significant risk factor for infection with *T. gondii*.³¹

In the present study, consumption of vegetables treated with untreated water was also identified as a risk factor for seropositivity. Oocysts lose their infectivity when water is heated

TABLE 3
Bivariate analysis of eating and drinking habits associated with infection with *Toxoplasma gondii* in 963 pregnant women in Fortaleza, Brazil*

Risk factor	No.	No. (%) IgG positive	Odds ratio (95% confidence interval)	P
Consumption of chicken				
Yes	926	637 (68.8)	1.19 (0.55–2.48)	0.593
No	37	24 (64.9)		
Consumption of raw or undercooked chicken				
Yes	15	10 (66.7)	0.91 (0.28–3.44)	1.0
No	947	650 (68.6)		
Consumption of chicken > 2 times/week				
Yes	508	370 (72.8)	1.52 (1.14–2.01)	0.003
No	454	290 (63.8)		
Consumption of pork				
Yes	290	196 (67.6)	0.93 (0.69–1.27)	0.650
No	673	465 (69.1)		
Consumption of raw or undercooked pork				
Yes	5	4 (80.0)	1.83 (0.18–90.67)	1.0
No	957	656 (68.8)		
Consumption of pork > 2 times/week				
Yes	20	16 (80.0)	1.84 (0.59–7.66)	0.336
No	943	645 (66.4)		
Tasting of meat while cooking				
Yes	348	242 (69.9)	1.10 (0.82–1.49)	0.515
No	615	417 (67.8)		
Consumption of milk				
Yes	813	548 (67.4)	0.68 (0.44–1.02)	0.056
No	150	113 (75.3)		
Consumption of powder milk				
Yes	631	419 (66.4)	0.73 (0.54–0.99)	0.04
No	330	241 (73.0)		
Consumption of milk > 2 times/week				
Yes	613	400 (65.3)	0.62 (0.42–0.84)	0.001
No	344	259 (75.3)		
Consumption of cheese				
Yes	750	513 (68.0)	0.95 (0.67–1.33)	0.802
No	213	148 (69.5)		
Consumption of cheese > 2 times/week				
Yes	220	142 (64.5)	0.78 (0.57–1.10)	0.138
No	743	519 (69.9)		
Consumption of dindin				
Yes	632	452 (71.5)	1.46 (1.09–1.96)	0.009
No	331	209 (63.1)		
Consumption of dindin > 2 times/week				
Yes	312	221 (70.8)	1.16 (0.86–1.59)	0.3
No	649	438 (67.5)		
Consumption of raw vegetables				
Yes	751	518 (69.0)	1.07 (0.76–1.50)	0.676
No	212	143 (67.5)		
Washing vegetables with untreated† water before eating				
No	349	256 (73.4)	1.42 (1.06–1.92)	0.017
Yes	604	398 (65.9)		
Consumption of vegetables > 2 times/week				
Yes	480	318 (66.3)	0.81 (0.61–1.07)	0.144
No	476	337 (70.8)		
Drinking treated water				
No	196	145 (74.0)	1.38 (0.96–2.00)	0.084
Yes	765	515 (67.3)		

* Dindin = ice made of water, fruits, and sugar.

† Untreated water = water unfiltered or not boiled.

to 60°C for at least one minute.³² Other studies in Brazil demonstrated that drinking untreated water was a significant risk factor for infection, also in outbreak situations.^{24,31} Water as a source of infection likely plays a more important role than ingestion of raw meat in northeastern Brazil. In light of these results, different routes of oral transmission by contaminated water, including the role of water ice, should be further investigated.

We identified an association of dogs, rather than cats, with *T. gondii* infection. Dogs may act as mechanical vectors by rolling in foul-smelling substances and ingesting fecal material.³³

The lack of an association of cat ownership is surprising. Stray cats represent a severe risk for toxoplasmosis in Brazil because up to 87% of stray cats in Brazil are infected with *T. gondii*.^{34,35} Cats often spread oocysts outside the home, and feral cats may be responsible for much of the environmental contamination with oocysts because sporulated oocysts maintain infectivity for a long time in water or soil.^{1,36}

Our study has several limitations. First, the low incidence of IgM against *T. gondii* enabled us to investigate risk factors only in IgG-positive pregnant women and may thereby have failed to identify direct causal associations between infection

TABLE 4

Multivariate logistic regression analysis of risk factors for infection with *Toxoplasma gondii* in 963 pregnant women in Fortaleza, Brazil (age-adjusted)*

Variable	Odds ratio (95% confidence interval)	P
Consumption of homemade water ice (dindin)	1.49 (1.09–2.04)	0.01
Consumption of chicken > 2 times/week	1.49 (1.12–2.00)	0.007
Dog ownership	1.46 (1.07–1.98)	0.02
Consumption of vegetables washed with untreated water	1.43 (1.05–1.94)	0.03
Monthly income ≤ 1 MMW	1.40 (1.02–1.90)	0.04

* Dindin = ice made of water, fruits, and sugar; MMW = monthly minimum wage (U.S. \$120).

and risk factors of importance. Second, detection of IgG but not IgM against *T. gondii* defines the classical serologic pattern of latent infection. However, a negative IgM titer in the third trimester does not necessarily rule out an acute infection in early pregnancy. Third, the rather short study period may have failed to detect seasonal or other differences in risk factors. However, we do not expect seasonal differences to be of importance because of the relatively stable climate in this region. Fourth, the results were obtained in a population of pregnant women in one academic medical center in Fortaleza and should be carefully generalized to pregnant women living in Fortaleza and its metropolitan region.

Results of the present study therefore advocate implementation of primary prevention for young persons with a special focus on the population with a low socioeconomic status. Trained community health agents of the Brazilian Public Healthcare System (Family Health Program/*Programa de Saúde da Família*) may provide information to families about risks and prevention measures for infection with *T. gondii*. Such health education should reach the whole population at risk, and must be accurate, practical, consistent, and clear.³⁷

In conclusion, we have shown that the seroprevalence of IgG against *T. gondii* is high among pregnant women in north-eastern Brazil. Young women with a low socioeconomic status who consume food and drinks prepared with water are at highest risk for acquiring this infection.

Received February 8, 2010. Accepted for publication April 19, 2010.

Acknowledgments: We thank the pregnant women for their participation in the study, and the physicians, nurses, and healthcare providers in Fortaleza, especially those from University hospital MEAC for their collaboration with our research team. Some of the results in this report are included in the medical theses of Susann Sroka and Andreas Winter.

Financial support: Jörg Heukelbach is research fellow from the Conselho Nacional de Desenvolvimento Científico e Tecnológico/Brazil. Susann Sroka, Nina Bartelheimer, and Andreas Winter were supported by grants from the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior/Deutscher Akademischer Austausch Dienst Integrated Projects in University Co-operation between Brazil and Germany academic exchange program.

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