

Seroprevalence and risk factors of *Toxoplasma gondii* infection among pregnant women in Trinidad and Tobago

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ABSTRACT

Objective. To determine the seroprevalence of toxoplasmosis and the frequency of risk factors for the disease in women attending antenatal clinics in hospitals and local health centers in Trinidad and Tobago.

Methods. From November 2002–September 2003, 232 pregnant women at the antenatal clinics of two large hospitals were sampled during their first trimesters. From October 2003–February 2005, 218 pregnant women at five health care centers were followed through three trimesters, wherever possible, and blood samples collected. Sera were screened for *Toxoplasma gondii* IgG and IgM immunoglobulins using an enzyme immunoassay. Data on demographics, as well as practices considered to be risk factors for toxoplasmosis, were obtained.

Results. Of the 232 women sampled at the two hospital clinics, 83 (35.8%) and 8 (3.4%) were seropositive for immunoglobulins of *T. gondii* IgG and IgM, respectively. Of the 218 women at the health centers, 76 (34.9%) had evidence of past infection (IgG), while 26 (11.9%) were seropositive for IgM immunoglobulin, suggesting new infections during pregnancy. Only having “3 or more” children was significantly associated with infection by *T. gondii*.

Conclusion. In Trinidad and Tobago, the seroprevalence of past infection by *T. gondii* in pregnant women is relatively low (39.3%) for patients in both hospital and health center clinics. Consequently, there is an elevated risk of primary infection during pregnancy and the potential for congenital infection.

Key words

Toxoplasmosis, congenital; prenatal care; Trinidad and Tobago.

Toxoplasma gondii is a well known parasitic zoonosis that is a causative

agent for abortions, stillbirths, eye problems, and mental retardation in the children of women who acquire primary infection during pregnancy (1–3). Cats and other members of the Felidae are the definitive hosts of the parasite and shed the oocysts after they are infected (3–5). These oocysts contain infective sporozoites that can then cause human infection by fecal-oral transmission. Humans can also be exposed to

bradyzoites, contained in tissue cysts of the intermediate hosts—particularly food animals—through consumption of improperly cooked meat and meat products or water (3, 6–8).

To prevent the clinical consequences in exposed children, some nations adopted prophylactic treatment of seropositive women (9–11) and neonatal screening (12, 13). Seroprevalence rates for past infections in pregnant

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women as detected by *T. gondii* IgG immunoglobulins have varied from 0.79% to 85% in various countries (14–17). For newly acquired *T. gondii* infections (IgM) during pregnancy, seroprevalence rates have varied from 0.2% to 25.7% across countries (13, 18–21). Clinical toxoplasmosis in children has also been reported with a frequency ranging from 0.02% to 0.5% (18, 22, 23).

In the English-speaking Caribbean, there is a dearth of information on congenital toxoplasmosis. Reports exist on toxoplasmosis in antenatal women in Trinidad (43.3%), pregnant women in Jamaica (57%), and young Jamaican women (45%) (24–26). In all these studies, only past infections were documented. To date, the only reported case of clinical toxoplasmosis in the area was documented almost 50 years ago in Jamaica (27). More recently, Adesiyun et al. (28) have been the first to demonstrate the serological evidence of congenital toxoplasmosis in Trinidad and Tobago, and possibly the English-speaking Caribbean, where 0.4% of cord blood samples tested were seropositive for *T. gondii* IgM immunoglobulin.

Because certain risk factors have been associated with toxoplasmosis (29, 30) and the demographics of women attending hospital versus local health center clinics (LHC) differ, we performed a descriptive and cross-sectional study to compare *T. gondii* seroprevalence rates during the first trimester of pregnancy at both hospital and health center clinics. In addition, at the LHCs, we conducted a longitudinal cohort study following women during their antenatal visits and taking one blood sample in each of the trimesters whenever practicable.

MATERIALS AND METHODS

Study population

From November 2002–September 2003, we studied a total of 156 women attending the Port-of-Spain General Hospital (POSGH) and 76 women registered at the Mount Hope Maternity

Hospital (MHMH), two of the three largest health institutions on the island. Both of these hospitals are located in the East-West Corridor of Trinidad. The patients attending the antenatal clinics of these two hospitals come from all over the island to consult with the specialists from the University of the West Indies that serve there. The patients cover the spectrum of socioeconomic, from low to high.

In addition, from October 2003–February 2005, we studied a total of 218 women attending the antenatal clinics at five LHCs (Barataria, El Socorro, St. Joseph, Success Laventille, and Upper Laventille) in the East-West Corridor of the island of Trinidad. One of the demographic characteristics of the population served by these centers is that a majority belong to the low- to medium-socioeconomic group.

Participant selection criteria

The study was designed to include all pregnant women who expressed an interest in participating in both the cross-sectional and cohort aspects of the investigation during the given periods. For the cohort study, it was anticipated that there would be some attrition beyond the control phase. The design was also not to determine the overall number of pregnant women who presented at the hospitals or health centers during the study period.

Sociodemographic data

Upon recruitment of women into the study, a questionnaire was administered to elicit data on demographics and practices considered to be risk factors for toxoplasmosis. The demographic data included age, gender, marital status, race, religion, occupation, level of education, and number of children. The toxoplasmosis risk-factor questions centered on the presence of cats in the household, handling of cat litter, practice of outdoor gardening, consumption of rare/improperly cooked meat and unwashed vegetables, and working on a farm.

Sample collection

For each sample, approximately 5 mL of blood was aseptically drawn by venipuncture into a tube without anticoagulant and refrigerated overnight at 4°C. It was then centrifuged, serum harvested, and stored at –20°C until tested.

The design of the longitudinal health center study required that each pregnant woman be sampled once each trimester. Due to attrition, a number of the patients could only be sampled twice (first and second trimester), or once (first, second, or third trimester).

Toxoplasma gondii IgG and IgM immunoglobulins assay

Enzyme immunoassay (EIA) test kits (Diamedix, Miami, Florida, United States of America), both IgG and IgM kits, were used to detect *T. gondii* immunoglobulins in serum samples as earlier described (28).

Ethics

The ethics committees of the regional health authorities responsible for the five health centers and those of both hospital clinics approved the study prior to commencement. The objectives and protocol for the study were explained to all the participants and a written consent was obtained from each.

Statistical analysis

The risk factors associated with exposure to toxoplasmosis, such as age, race, presence of cats in household, handling cat litter, outdoor gardening, etc., were tested for significance using the Chi-square test for independence, as well as by determining the relative risk and odds ratio for the cohort study and cross-sectional study, respectively. All tests were two-tailed and interpreted at the 5% confidence level of significance. Data were processed using the Statistical Package

for Social Sciences, version 10 (SPSS Inc., Chicago, Illinois, United States).

RESULTS

Overall, of the 450 pregnant participants at both the hospitals and the health center clinics, 177 (39.3%) tested in the first trimester were positive for IgG antibodies, while a total of 34 (7.6%) were positive for IgM antibodies. Of the 232 patients at hospital clinics, 83 (35.8%) and 8 (3.4%) were seropositive for *T. gondii* IgG and IgM immunoglobulins, respectively. For the 76 pregnant women in their first trimester sampled in MHMH antenatal clinic, 33 (43.4%) were positive for *T. gondii* IgG immunoglobulin and 4 (5.3%) for IgM immunoglobulin, compared with 53 (33.9%) and 4 (2.6%), respectively, of 156 women sampled at the POSGH clinic. The difference between hospitals was not statistically significant ($P > 0.05$).

Of the 218 women sampled in local health centers, 76 (34.9%) were seropositive for *T. gondii* IgG immunoglobulin at least once during the pregnancy; however, 23 (10.6%) were positive for *T. gondii* IgM. Table 1 shows the seroprevalence of both *T. gondii* IgG and IgM immunoglobulins across the five health centers studied and for the three groups of samplings. The seroprevalence of IgG immunoglobulin varied significantly ($P > 0.05$) across health centers.

The seroprevalence of toxoplasmosis in women with various demographic backgrounds is shown in Table 2. Of all the demographic and risk factors studied, a statistically significant difference ($P > 0.05$) was found only between women with three or more children and those with lower parity. Those with three or more children had a higher seroprevalence rate of toxoplasmosis. Differences in seroprevalence for *T. gondii* infections were not statistically significant ($P > 0.05$) for educational status and occupation between the two groups (Table 3). In addition, association with cats, outdoor gardening, and/or farming were not significant ($P > 0.05$) to seropositivity in samples taken at both hospital clinics and health centers.

DISCUSSION

The finding that only 39.3% of the 450 total women sampled at the antenatal clinics in the country had been exposed to *T. gondii* by the first trimester is considerably lower than the 60.4% seropositive rate reported for Turkey (19), 74.5% for Brazil (31), and 75.4% for Nigeria (32). In Trinidad, Orrett (24), in a cross-sectional study, reported that 43.3% of women sampled at health centers during their first trimester of pregnancy were seropositive for *T. gondii* infection. The 39.3% seroprevalence rate detected for past exposure in the current study, al-

though health-facility based as opposed to population-based, demonstrates a decreasing seroprevalence compared with published data in the Caribbean (24–26), a finding consistent with trends across Europe and North America (1, 3, 18, 33). The implication is that a high percentage of women of child-bearing age in Trinidad and Tobago have a high risk of contracting primary *T. gondii* infection during pregnancy, with the associated clinical manifestations in children and young adults (1, 3, 34). It is known that pregnant women with pre-existing infection by the pathogen, often acquired during childhood, are unlikely to transmit the infection to their fetuses during pregnancy due to the long-term persistence of IgG (5, 34). Serological evidence of *T. gondii* infection exists in slaughter animals (35), dogs (36), and cats in Trinidad (Watkins and Abdool, unpublished data).

It is important that serological evidence of potential congenital infection in unborn children was suggested by the detection of *T. gondii* IgM immunoglobulin in 3.4% or 8 of the 232 women sampled at hospital clinics and 10.6% (range of 2.0%–21.8%) or 23 of the 218 women longitudinally tested at the health centers. It has been established that acquisition of primary infection during pregnancy, as detected by seroconversion (i.e., seronegative at one trimester and becoming seropositive (IgG) in a subsequent trimester) or production of IgM,

TABLE 1. Frequency of toxoplasmosis in pregnant women sampled at health centers, in all three trimesters, only first and second trimesters, or only first trimester, Trinidad and Tobago

| Health centers | All three trimesters | | | First and second trimester only | | | First trimester only | | |
|--------------------|----------------------|-----------------------|-----------|---------------------------------|-----------------------|-----------|----------------------|-----------------------|---------|
| | No. of patients | No. (%) positive for: | | No. of patients | No. (%) positive for: | | No. of patients | No. (%) positive for: | |
| | | IgG | IgM | | IgG | IgM | | IgG | IgM |
| Barataria | 10 | 4 (40.0) | 3 (30.0) | 18 | 6 (33.3) | 2 (11.1) | 32 | 11 (34.4) | 0 (0.0) |
| El Socorro | 26 | 8 (30.8) | 4 (15.4) | 14 | 3 (21.4) | 3 (21.4) | 23 | 7 (30.4) | 0 (0.0) |
| St. Joseph | 13 | 4 (30.8) | 3 (23.1) | 12 | 3 (33.3) | 3 (25.0) | 16 | 8 (50.0) | 1 (6.3) |
| Success Laventille | 8 | 3 (37.5) | 1 (12.5) | 7 | 3 (42.9) | 1 (14.3) | 26 | 12 (46.2) | 0 (0.0) |
| Upper Laventille | 4 | 1 (25.0) | 1 (25.0) | 4 | 1 (25.0) | 1 (25.0) | 5 | 2 (40.0) | 0 (0.0) |
| Total | 61 | 20 (33.3) | 12 (19.7) | 55 | 16 (29.1) | 10 (18.2) | 102 | 40 (39.2) | 1 (1.0) |

TABLE 2. Seroprevalence of *Toxoplasma gondii* immunoglobulins (IgG and IgM) by mother-related risk factors, Trinidad and Tobago

| Risk factor | Hospital clinics | | | Health center clinics | | |
|----------------------------|---------------------|---|----------------|-----------------------|---|----------------|
| | No. of women tested | No. (%) positive for IgG/IgM ^a | <i>P</i> value | No. of women tested | No. (%) positive for IgG/IgM ^b | <i>P</i> value |
| Age (years) | | | | | | |
| <20 | 55 | 16 (29.1) | 0.174 | 62 | 21 (33.9) | 0.575 |
| 21-30 | 105 | 39 (37.1) | | 127 | 32 (25.2) | |
| 31-40 | 68 | 30 (44.1) | | 23 | 8 (34.8) | |
| 41-50 | 2 | 1 (50.0) | | 1 | 1 (100.0) | |
| Not available | 2 | 2 (100.0) | | 5 | 3 (60.0) | |
| Marital status | | | | | | |
| Single | 76 | 24 (31.6) | 0.611 | 73 | 23 (31.5) | 0.08 |
| Married | 68 | 28 (41.2) | | 58 | 17 (29.3) | |
| Other ^c | 88 | 30 (34.1) | | 87 | 25 (29.0) | |
| Religion | | | | | | |
| Christian | 200 | 74 (37.0) | 0.669 | 176 | 51 (29.0) | 0.481 |
| Hindu | 18 | 7 (38.9) | | 26 | 8 (30.8) | |
| Islam | 10 | 5 (50.0) | | 8 | 4 (50.0) | |
| Other | 4 | 2 (50.0) | | 8 | 2 (25.0) | |
| Race | | | | | | |
| African | 106 | 33 (31.1) | 0.196 | 96 | 27 (28.1) | 0.836 |
| East Indian | 38 | 16 (42.1) | | 47 | 15 (31.9) | |
| Mixed | 86 | 38 (44.2) | | 73 | 21 (28.8) | |
| Other | 2 | 1 (50.0) | | 2 | 1 (50.0) | |
| No. of children | | | | | | |
| None | 123 | 34 (27.6) | 0.002 | 110 | 30 (27.3) | 0.756 |
| One | 48 | 25 (52.1) | | 64 | 20 (31.3) | |
| Two | 26 | 10 (38.5) | | 29 | 8 (27.6) | |
| Three | 18 | 12 (66.7) | | 6 | 2 (33.3) | |
| More than three | 17 | 7 (41.2) | | 9 | 5 (55.6) | |
| Experience of miscarriage | | | | | | |
| Yes | 66 | 23 (34.8) | 0.376 | 39 | 13 (33.3) | 0.568 |
| No | 165 | 64 (38.8) | | 92 | 31 (33.7) | |
| Not applicable/no response | 1 | 1 (100.0) | | 87 | 21 (24.1) | |
| Experience of stillbirths | | | | | | |
| Yes | 6 | 3 (50.0) | 0.36 | 4 | 1 (25.0) | 0.694 |
| No | 225 | 84 (37.3) | | 126 | 43 (34.1) | |
| Not applicable/no response | 1 | 1 (100.0) | | 88 | 21 (23.7) | |

^a Positive for IgG or IgM or both among 232 women.

^b Positive for IgG or IgM or both among 218 women.

^c Divorced, separated, or common-law relationship.

has the potential to cause congenital infection in children and clinical symptoms, such as abortion, stillbirths, mental retardation, and eye problems (1, 18, 34). The rate of detection of IgM antibodies (7.6%) found in the 450 women in this study is considerably higher than what has been reported in Brazil, 0.64% (20); Hungary, 0.5% (37); Slovenia, 0.6% (38); or the United States, 0.1% (33). The detection rate of IgM antibodies during pregnancy is also considerably higher in Trinidad than the 0.2%–2% reported

for Brazil, considered the highest in the world (21, 39). The frequency of *T. gondii* IgM immunoglobulin detected in this study could not be attributed to old infection—a possibility suggested by Gras et al. (40) who declared that in up to 27% of pregnant women IgM immunoglobulin levels persist for more than two years, making it difficult to pinpoint the timing of infection. However, this is an unlikely scenario because in the longitudinal component of this study, with a total of 116 women sampled at each of the

three trimesters or during each of the first and second trimesters, 22 (19.0%) were seropositive for IgM for the first time during their pregnancies. It is pertinent, however, to mention that one cannot always equate the detection of *T. gondii*-specific IgM to acute infection since commercial test kits have yielded false-positives (41).

Although cats are the primary source of oocysts that cause human and livestock infections (3, 5, 30), in our study, household cats or handling of cat litter were not found to be signif-

TABLE 3. Seroprevalence of *Toxoplasma gondii* immunoglobulins (IgG and IgM) in pregnant women by association with cats and other practices, Trinidad and Tobago

| Risk factor | Hospital clinics | | | Health center clinics | | |
|-------------------------------------|---------------------|---|----------------|-----------------------|---|----------------|
| | No. of women tested | No. (%) positive for IgG/IgM ^a | <i>P</i> value | No. of women tested | No. (%) positive for IgG/IgM ^b | <i>P</i> value |
| Cat in the household | | | | | | |
| Yes | 42 | 16 (38.1) | 0.379 | 33 | 11 (33.3) | 0.163 |
| No | 189 | 71 (37.6) | | 182 | 52 (28.6) | |
| No response | 1 | 1 (100.0) | | 3 | 1 (33.3) | |
| Handling of cat litter ^c | | | | | | |
| Yes | 2 | 1 (50.0) | 0.936 | 5 | 1 (20.0) | 0.473 |
| No | 40 | 15 (37.5) | | 28 | 12 (42.9) | |
| Practice outdoor gardening | | | | | | |
| Yes | 43 | 20 (46.5) | 0.183 | 36 | 10 (28.7) | 0.309 |
| No | 188 | 67 (35.6) | | 180 | 53 (29.4) | |
| Not applicable/no response | 1 | 1 (100.0) | | 2 | 1 (50.0) | |
| Work on farm | | | | | | |
| Yes | 10 | 6 (60.0) | 0.145 | 4 | 0 (0.0) | 0.191 |
| No | 221 | 81 (36.7) | | 212 | 63 (29.7) | |
| Not applicable/no response | 1 | 1 (100.0) | | 2 | 1 (50.0) | |

^a Positive for IgG or IgM or both among 232 women.

^b Positive for IgG or IgM or both among 218 women.

^c Of households with cats.

icantly associated with *T. gondii* infection in pregnant women. The finding correlates with an earlier report in Trinidad and Tobago (28) and elsewhere (6, 42), but differs from the findings of others (3, 4, 30). Similarly, outdoor gardening and farm work did not significantly affect occurrence of toxoplasmosis, contrary to reports by others that these are independent risk factors for the infection (6, 14, 35). In our study, although age was not found to be significantly associated with toxoplasmosis as earlier observed (17), women with three children sampled in hospital clinics, were found to have significantly higher seroprevalence of *T. gondii* infection compared with other women, a finding that agrees with those of others (17, 43). Overall, in both hospitals and health centers, failure to detect significant association between most of the risk factors studied may reflect the fact that few antenatal women (only 34 women) acquired primary *T. gondii* (IgM) infections during pregnancy, making it difficult to meaningfully compare the frequency of risk factors in this subpopulation with the antenatal women who were

seronegative for *T. gondii* infection (303 women), i.e., negative for both IgM and IgG.

A few of limitations of the study design include the fact that some attrition was experienced in the cohort study, and the participants lost may have had different outcomes than those who were not lost. Another limitation is that the design did not consider the total number of pregnant women who presented at the health centers during the study period, compared to the number who agreed to participate. It is felt, however, that these limitations did not significantly affect the findings.

This study revealed a high percentage of pregnant women seronegative for toxoplasmosis in the first trimester and an unexpectedly high rate of possible primary infection during pregnancy due to detection of IgM immunoglobulin, with its potential for congenital toxoplasmosis. In this context, it is of concern that no antenatal screening, educational programs, or treatment of seropositive pregnant women is currently practiced. The clinical impact of toxoplasmosis is also

not known in this population. A combination of these approaches has been employed in a cost-effective way in countries with varying seroprevalence of toxoplasmosis (9, 13, 30, 33). Taking into account that prenatal screening may be a viable option for populations with high rates of primary infection during pregnancy (up to 3.5%) (44), it is worth considering routine screening as an interim measure, with prompt treatment for those who become infected during pregnancy. It is relevant to mention that contrary to reports suggesting evidence for prenatal treatment of toxoplasmosis (9, 34, 45), there are reports that show a lack of support for this approach (45–47).

In conclusion, in Trinidad and Tobago the potential for congenital infection is high and follow-up studies of infected children are needed to assess the clinical implications of infection. To reduce exposure of pregnant women to this disease, it is imperative that educational programs be mounted to create awareness among the public, as well as among health personnel, and to work towards prenatal screening for toxoplasmosis.

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RESUMEN

Seroprevalencia y factores de riesgo de la infección por *Toxoplasma gondii* en mujeres embarazadas en Trinidad y Tobago

Objetivo. Determinar la seroprevalencia de toxoplasmosis y la frecuencia de los factores de riesgo de la enfermedad en mujeres que asisten a consultas prenatales en hospitales y centros de salud en Trinidad y Tobago.

Métodos. Entre noviembre de 2002 y septiembre de 2003 se tomaron muestras de 232 mujeres que asistieron a consultas prenatales en dos grandes hospitales durante su primer trimestre de embarazo. Entre octubre de 2003 y febrero de 2005 se realizó el seguimiento durante tres trimestres, siempre que fue posible, y se tomaron muestras de sangre de 218 embarazadas atendidas en cinco centros de salud. Se analizaron las muestras de suero para detectar anticuerpos de las clases IgG e IgM contra *Toxoplasma gondii* mediante ensayos inmunoenzimáticos. Se tomó nota de los datos demográficos y las prácticas consideradas factores de riesgo para la toxoplasmosis.

Resultados. De las 232 mujeres estudiadas en los dos hospitales, 83 (35,8%) y 8 (3,4%) resultaron seropositivas para anticuerpos IgG e IgM contra *T. gondii*, respectivamente. De las 218 mujeres estudiadas en los centros de salud, 76 (34,9%) presentaban evidencias de infección pasada (anticuerpos IgG), mientras 26 (11,9%) eran positivas a anticuerpos IgM, lo que indicaba una infección reciente durante el embarazo. El único factor asociado significativamente con la infección por *T. gondii* fue tener tres hijos o más.

Conclusiones. En Trinidad y Tobago, la seroprevalencia de infección pasada por *T. gondii* encontrada en mujeres embarazadas que se atendían en los hospitales y los centros de salud fue relativamente baja (39,3%). Por consiguiente, existe un elevado riesgo de infección primaria durante el embarazo con la posibilidad de infección congénita.

Palabras clave

Toxoplasmosis congénita, atención prenatal, Trinidad y Tobago.