Seroepidemiology of *Toxoplasma* Infection in Pregnant Women in Qom Province, Iran (2010)

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Abstract: Background: *Toxoplasma gondii* is a protozoan parasite that can infect a wide range of hosts, including humans. Infection with *T. gondii* is potentially life threatening in immunocompromised individuals. Moreover, the infection can be detrimental during pregnancy, often leading to abortion of the fetus. The purpose of this work was to evaluate the seroepidemiology of *T. gondii* infection among Qom's pregnant women in 20l0. **Materials and methods**: In this descriptive, cross-sectional study, 200 serum samples were selected randomly and examined for toxoplasmosis by ELISA and IFA methods. The relationship between seropositivity for toxoplasmosis on the one hand, and some important factors on the other hand, were evaluated. **Results**: Total prevalence of Anti-*Toxoplasma* was 45.3%. According to the data reported by patients, toxoplasmosis had direct relationship with residential areas, consumption of semi-cooked meat, consumption of unwashed raw vegetables, and being in contact with cats. There is a statistically significant relationship between age and occurrence of the infection with regard to IgG seropositivity. However, such relationship could not be found for IgM. The highest infection rate was observed in the age range of 2l to 25. **Discussion**: Education about the routes of the disease transmission and performing *toxoplasma* test before pregnancy can be effective reducing the prevalence rate. [Lame Akhlaghi, Shahnaz shirbazou, Fatemeh Maleki, Alireza Keyghobadi, Yaser tabaraei, Fatemeh Tabatabaie**, Seroepidemiology of *Toxoplasma* Infection in Pregnant Women in Qom Province, Iran (2010)**, *Life Sci J* 2013;10(7s): 322-325] (ISSN:1097-8135). http://www.lifesciencesite.com.

Introduction

Toxoplasma gondii is an intracellular parasite that infects a wide variety of hosts, including humans. Human infection occurs through two main routes, ingestion of undercooked meat containing cysts of the parasite and ingestion of oocysts passed into the environment by cats. *T. gondii* infects a large proportion of the world's population. Individuals at risk include fetuses, newborns, and immunologically compromised individuals. If a pregnant woman contacts with Toxoplasma, the parasite may passed through the placenta to the fetus, resulting in congenital toxoplasmosis, which is a cause of fetal mortality and malformation. Therefore, if mother is infected by *Toxoplasma* during pregnancy, the infection could be transferred to fetus. However, if mother is infected before pregnancy, the infection cannot be transferred to the fetus, and the mother acquires immunity against future infections of *Toxoplasma*. Several studies have been conducted on seroepidemiology of toxoplasmosis during pregnancy and on the women within the childbearing age in Iran and other countries. The mean prevalence of toxoplasmosis in different areas of Iran has been reported to be about 35%. The rates reported indicate that the prevalence rate differs from one location to another in different parts of Iran and the world, and even within one country with different conditions. However, in recent years there is not any report of the prevalence rate of *Toxoplasma* infection in Qom, Iran (1, 2, 3). Thus, there is a need to carry out a study on pregnant women in Qom to estimate the burden of toxoplasmosis in this area.

Materials and methods

In this cross-sectional study, the samples were randomly obtained from women who referred to Obstetrics state hospital of Qom for routine pregnancy test, and the test result was positive. The sample size was 200, with the d value of 0.04 at the confidence level 96%, based on the infection prevalence rate of 35%. Blood samples were collected and the sera separated by centrifugation at 3000 rpm for 5 min and frozen at -20 °C until use. In indirect fluorescence antibody (IFA) test, antigen was prepared from tachyzoites of *T. gondii* RH strain and inoculated in peritoneum of Balb/c mice. After four days, tachyzoites were collected by peritoneal

washing and centrifuged at 2000 rpm, washed three times with PBS, coated on microscopic slides, and frozen at -20 °C until use. Sera were diluted serially and *Toxoplasma* (*IgG* or IgM) antibodies were detected with indirect immunofluorescent antibody method in titers of 1:10. In this study, titers higher than 1:10 were considered positive. Enzyme-linked immunosorbent assay (ELISA) test was used to screen all the samples for *T. gondii* antibodies according to the manufacturer's guidelines. Testing was carried out in batches. Briefly, 96-well microtiter polystyrene plates were sensitized with sonicated *T. gondii* antigen provided with the kit at a concentration of 100 μg/ml in carbonate buffer (pH 9.5) and blocked with bovine fetal serum to 1% in PBS-Tween 20 (0.01%). The sera were diluted 1:100 using the assay diluent supplied with the kit and incubated at 25 °C for 30 min, and rinsed four times with wash buffer. Then, 100 μl of the detecting anti-IgG conjugate or anti-IgM conjugate was added to each well. Plates were incubated humidified at 37 °C for 30 min and were then washed five times before addition of 100 μl of tetramethylbenzidine substrate. The plates were incubated for 10 min at room temperature in the dark, and the colorimetric reaction was stopped by adding 100 μl of stopping reagent per well. The absorbance of the samples and controls was determined at 450 nm by ELISA microplate reader. Positive and negative control sera were included in each run. The results were compared with cutoff and expressed in IU/ml by quantitative estimation using calibration curve constructed with cutoff and three positive controls (an index value of < 0.253 was considered negative for anti-*Toxoplasma* antibody, a value of ≥ 0.253 but ≤ 0.343 is considered equivocal, and a value of > 0.343 is considered positive). Equivocal results were not included in the analyses. Ethical Committee of the University approved this study. Data on age, contact with cat, habit of ingestion undercooked meat or unwashed vegetables, and area of residence were recorded in a questionnaire (4, 5). We used the SPSS 11.5 software for analyzing the data. In order to check for statistic difference, chi-square test and Student's *t*-test were adopted. Differences between two groups were considered significant when p values were < 0.05.

Results

The average rate of *Toxoplasma* prevalence was 45.3% by the two serological methods used. The results obtained from the two methods had some minor difference; however, the differences were not statistically significant. Only six cases (%1) were positive for Toxoplasma with the ELISA method, but negative in the IFA. In diagnosis of toxoplasma, ELISA method is preferred to IFA, owing to its high

sensitivity and specificity, simple application, and its low costs. The results indicated that women who were at risk should consult with an obstetrician. Among the 200 serum samples analyzed, 87 samples were found to be positive for anti-*T. gondii* IgG and IgM, corresponding to an overall prevalence of 45.3%. Mean result obtained from the two methods showed that most positive samples (76 cases or 39.6%) were positive for IgG, which indicates the chronic cases of toxoplasma infection, and a low number of cases (5.7% or 11 cases) were positive for IgM, which indicates the acute cases. The highest infection rate was observed in the age range of 21 to 25. Regarding the IgG test result, a statistically significant relationship was found between age and the rate of infection. However, such relationship could not be established for IgM. Among the pregnant women who were in contact with cats, 65% and 25% were positive for IgG and IgM, respectively. Moreover, 40% and 30% of the people who lived in urban and suburbs were positive for IgG, respectively. The rates for IgM antibody were 70% and 15%, respectively. Moreover, among the pregnant women who used semi-cooked meat and unsterile row vegetables, 70% and 25% were positive for IgG and IgM, respectively. The results indicated that there is a statistically significant relationship between being infected with *Toxoplasma* on the one hand, and using semi-cooked meat, using unsterile row vegetables, having contact with cats, and the residence place on the other hand. The most frequent signs and symptoms observed in patients were fever, jaundice, muscle pain, GI and minor discomforts, skin blisters and rashes, chilling, and lymphadenitis.

Discussion

Toxoplasma gondii infection is a growing worldwide health concern, with estimates of up to one third of the world's population infected with the parasite. The ubiquitous protozoan parasite *Toxoplasma gondii* is a major cause of morbidity and mortality in neonates and immunocompromised hosts (6, 7). Since currently available therapies are not fully effective against the infection, and no *T. gondii* vaccine is available, efforts to reduce toxoplasmosis transmission are crucial to reduce the impact of toxoplasmosis. Prevention of congenital toxoplasmosis in pregnant women has been based on serological tests for *Toxoplasma* antibodies (8).

Our study showed that 45.3% (87/200) of pregnant women were seropositive for the parasite. It was observed that the prevalence IgG antibody against *T. gondii* in the population studied was 39.6% (76/200), while the arte for IgM antibody was 5.7% (11/200).There was not any new report about prevalence of the infection in Qom, a city of west-

central Iran, located south-southwest of Tehran. In a study in Hamadan carried out by Rabiee et al., seroprevalence of *Toxoplasma* infection in women aged 15 to 45 was 38.9%. The previous studies showed that the seroprevalence of toxoplasmosis was 84%, 74.6%, 54.2%, 42.8%, and 45.5% in Tehran, Mazandaran, Kashan, Qom, and Karaj, respectively (9-14). In another study in Zurich, the seroprevalence of women within the childbearing age (20- 40) was 40%. The highest seroprevalence has been reported from France, again in women of reproductive age. Lower prevalence rates were found in North America or Oceania. Two different studies in general population showed that seroprevalence of the infection in western and eastern parts of the Caspian basin were 87% and 55.7%, respectively. In Europe, the highest rates were observed in the central and southern Europe, while the lowest rates were reported from the north Europe. The prevalence rates of the infection were reported to be 0.79%, 30.1%, and 31.7% in Korea, Turkey, and Jordan, respectively.

These differences may be attributed to climate condition, nutritional behavior, socioeconomic state, and keeping cats. Moreover, the differences observed between various countries could result from assay and sampling methods, as well as the geographic and other temporal factors. The effect of these factors can be evaluate dby carrying out studies at public health level, individual characteristics, personal habits, and geographical and regional conditions, and also comparison of different serological methods for evaluation of antibodies against *Toxoplasma.* Regarding the results obtained, about half of the pregnant women studied (54.7%) were at risk of infection with *T. gondii*; thus, preventive measures should be applied. There is a statistically significant relationship between age and the prevalence of the infection with regard to IgG. However, such relationship could not be found for IgM. The reason for the rise in antibody titers in higher ages is not clear. An explanation would be the increased cumulative exposure to the parasite in higher ages. Statistically significant relationships could be established between toxoplasmosis on the one hand, and the residential area, consumption of semi-cooked meat, taking unwashed raw vegetables, and being in contact with cats, as the main risk factors of the disease. Preventive measures against toxoplasmosis should be applied according to the infection seroprevalence, gestational age, and the health facilities available. Moreover, pregnant women should be aware of the transmission routes of the infection. In this regard, courses and seminars should be held by the organizations in charge of health services in the city (15-18).

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