# Seroprevalence of *Toxoplasmosis* in High School Girls in Fasa District, Iran

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#### ABSTRACT

Background: Toxoplasmosis is a parasitic disease caused by protozoan, Toxoplasma gondii. Infections of human are common and are usually asymptomatic. The infection may be serious if is transmitted to the fetus during pregnancy. Prophylactic measures, early detection of the infection and treatment can avoid congenital toxoplasmosis and many long term effects. **Objective:** Seroepidemiological study in young girls is useful to determine the prevalence of infection and to design prevention policies for them after marriage and during their pregnancy. This study was carried out in the years 2000-2001 in the region of Fasa of Fars province in the South of Iran, as a descriptive, analytic and cross sectional study. Methods: Serum Samples of 947 students were collected from high school girls of Fasa and studied by enzyme linked immunosorbant assay (ELISA). The positive and negative controls were also used. **Results:** The seroprevalance rate of toxoplasmosis ranged from 1 to 21 Percent in different parts of Fasa and 10% in all groups. Some variables including age, nutritional habits and contact with domestic cats were studied. Conclusion: The seroprevalence of toxoplasmosis in girls of various high schools of Fasa is different and it may be related to the level of hygiene in different parts of Fasa. Water and food contamination with cat stool in regions with high contact with domestic cats can play an important role in infection rates. People of such areas should eat well-cooked meat to reduce infection.

#### Keywords: Toxoplasmosis, Seroepidemiology, ELISA

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# INTRODUCTION

Toxoplasmosis is an infection caused by the intracellular protozoan parasite Toxoplasma gondii. It is found in humans worldwide and in mammals and birds as intermediate hosts. The cat is the main host of the parasite. Human infection results from ingestion of soil contaminated with cat litter, ingestion of raw or insufficient cooked meat (lamb, pork, and beef) and transmission from a mother to a fetus through the placenta (congenital infection) or by blood transfusion or organ transplantation. Most cases of primary infections are asymptomatic. The incubation period is 1 to 2 weeks. Congenital toxoplasmosis is caused by acute infection with Toxoplasma gondii in a pregnant woman for the first time. One-third of primary toxoplasmosis cases occurring during pregnancy lead to transplacental transmission and involvement of the fetus with pathological effects such as microcephaly, hydrocephaly, blindness, calcification of brain cells and even death in utero. Signs of congenital infection may be observed at birth or develop over the first few months of life and its severity depends on the duration of infection in pregnant women (1-3). The prevalence rate of the disease is different in various parts of the world and is related to various factors such as age, sociocultural and nutritional habits and contact with domestic cats. Seroepidemiological studies of female toxoplasmosis before delivering age will be very useful for designing preventional policies during child bearing age. 15-18 year old girls are suitable groups for such study. Premarital examinations are conducted to diagnose previously infected women from women who have not been previously infected (1-3). Toxoplasmosis sero-surveys have been held in many countries. Seropositivity in women of childbearing age in France, Germany, Belgium and Switzerland is as high as 37-58%, while in the Latin American countries such as Argentina, Brazil and Cuba is 51-72% (3,4). In Iran, seropositivity in high school girl students aged 15-19 y in Isfahan province in central part and Robatkarim district near Tehran were 17.5% and 17.7%, respectively and general seroprevalence was estimated to be about 51.8% (5,6).

# MATERIALS AND METHODS

A cross-sectional sero-survey of *Toxoplasma* IgG-antibodies in 947 high school girls was conducted. Sample groups were chosen from public high school girls aged between 14 to 19 years. Eleven girl public high schools were chosen. All of students appeared healthy. Written agreements were obtained from all participants or parents after explaining the purpose of the study. Questionnaire forms were filled by all participants or caretakers. Blood sampling was performed without anticoagulant according to standard techniques and after 30 min, the tubes were centrifuged at 2,000 rpm for 5 min and then sera were aliquoted in several labeled vials and kept frozen at -20°C. All serologic tests were performed after field work was done. Enzyme linked immunosorbent assay (ELISA) was done to measure IgG antibody (RADIM Toxo IgG Diagnostic kit, Italy). A serially diluted serum was poured in the *Toxoplasma* antigen-coated wells and incubated at 37 °C for 60 min. If anti-*T. gondii* antibody exists in sera of samples, it binds to the specific antigen, while unbound antibody and other serum proteins were removed by washing. Then 100µl of enzyme tracer was added to the microplate wells and incubated for 30 min at 37 °C. IgG-

antibody-Toxoplasma-antigen complexes attached to the microplate well and unbound conjugate were then washed. A chromogen solution containing tetramethylbenzidine with citrate-phosphate buffer and DMSO was then added and incubated at 37 °C for 10 min to develop the color and the reaction was stopped by an acid solution. The optical density was read at 450 nm by ELISA reader and converted into IU/mL of *T. gondii* IgG antibody through a standard curve. Each commercial microplate unit contained negative and positive standard control serums. Results lower than 6 IU/mL were considered negative for *T. gondii* IgG antibodies. Data were recorded and analyzed using SPSS 6.0 software. Once data consistency was checked out, frequencies and seroprevalence rates were calculated. Some variables including age, nutritional habits, contact with domestic cats and other pets and parents' education were also studied. The correlation between selected variables and seropositivity was analyzed by Chi square test. P < 0.05 was considered significant.

# RESULTS

From September 2000 to March 2001, blood samples of 947 high school girls aged between 14 to 19 years were analyzed for *T. gondii* IgG antibody using ELISA method. 96 cases were found to be seropositive. The seroprevalance rate of toxoplasmosis ranged from 1 to 21 percent in different parts of Fasa and averaged 10%. According to age, seropositive cases were highest in 16-year old group with an average of 30.2% (29 of 96 seropositive cases) and lowest in 19-year old group with an average of 3.12% (3 of 96 seropositive cases). Figure 1 shows the relationship between *T. gondii* seroprevalence and age. Among seropositive cases, about 60% had close contact with cats. Raw meat consumption was not observed and not well-cooked meat consumption was very rare in all studied groups. A relationship between *T. gondii* seroprevalence and the level of father's education was also studied. 40% (38 cases) of the fathers of seropositive girls were illiterate, 37% (35 cases) had average education and 23% (22 cases) had university education.



Figure 1. Relationship between *T. gondii* seroprevalence and age. As is indicated, the most seropositivity was at age 16.

# DISCUSSION

Seroepidemiological survey in different parts of the world indicates that the prevalence rates range from zero to 98% (7). Most studies are focused on childbearing age and

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pregnant women and also infants and immunodeficient patients (3,5,6,8). The highest infection rate has been reported from north provinces in Iran with a temperate and humid conditions while in Southern provinces with dry and warm climates, this rate decreases considerably. The main infection route of toxoplasmosis in Iran is through soil and water because in Iranian nutritional habits raw meat is not consumed (9). These study groups are representative of variable income groups attending public health and education facilities in Fasa. As a cross-sectionally designed study, our data refers only to prevalence rates, and incidence rates should be assessed through a separate study. Studies of toxoplasmosis seroprevalence have shown a statistical correlation with close contact with cats (2, 3). In our population a sigificant association was found between T. gondii seroprevalence and close contact with pets including cats in all studied groups, but not for each group alone. Of those groups which showed seropositivity of about 60%, had close contact with cats. Raw meat consumption was not observed and insufficiently cooked meat consumption was very rare in all study groups. Therefore no relationship could be found between raw meat consumption habits and T. gondii IgG-seropositivity in these groups. In geographical regions with raw meat consumption habits, there is a significant correlation between raw or undercooked meat consumption and increasing seroprevalence of toxoplasmosis (2, 3). A statistical association was found between *T. gondii* seroprevalence and the level of father's education. 40% of the fathers of seropositive girls were illiterate, 37% were of average education and 23% had university education. This study suggests that high T. gondii seroconversion, mostly before age 19, takes place in schools or homes through fecal-oral contamination by oocysts. The total prevalence rate was estimated to be 10% in our study groups. Fasa district in South of Iran is a warm and dry region and our results agree with investigation in Southern regions of Iran (9). Such studies on girls are very useful because premarital examinations are necessary to distinguish previously infected women from women who had not been previously infected. When a previously uninfected woman becomes pregnant, testing is programmed at her first prenatal examination. In addition, women are educated about prevention methods during pregnancy. Educating of women at childbearing age about minimizing their risk for infection is another approach to prevent toxoplasmosis. Educational interventions assume that increased knowledge results in awareness, which consequently results in changes in risky behavior and decline in infection rates. Health managers should emphasize the importance of avoiding raw or undercooked meat, handling raw meat safely, washing hands after gardening and preventing close contact with cats. (10)

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