



Seroprevalence of *Toxoplasma gondii* in the Rural Population of Qaemshahr, Northern Iran in 2019

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Abstract

Background: Toxoplasmosis is a worldwide infection caused by *Toxoplasma gondii*, an obligate intracellular protozoan. It infects approximately one-third of the world's population. This study was conducted to determine the anti-*T. gondii* antibodies (IgG and IgM) in the rural population of Qaemshahr, northern Iran, in 2019.

Methods: Serum samples of 350 individuals referred to the laboratories of the rural health centers in Qaemshahr, northern Iran, were collected from February to April 2019 and were assessed for anti-*T. gondii* antibodies (IgG and IgM) using the ELISA kits (Pishtazteb, Iran). The results were analyzed using the chi-square test.

Results: Totally, 296 (84.57%, 95% CI: 80.07%-88.2%) out of 350 serum samples were positive for anti-*T. gondii* IgG antibody. There was a significant correlation between seropositivity of anti-*T. gondii* IgG antibody and age ($P=0.000$), contact with soil ($P=0.000$), and job. None of the samples were positive for anti-*T. gondii* IgM antibody. No significant association was observed between the seroprevalence of *T. gondii* and the gender, contact with the cat and other animals, and ingestion of undercooked meat, non-chlorinated water, undercooked egg, raw milk and vegetables.

Conclusions: Results of the study showed that the prevalence of toxoplasmosis was much higher among the rural individuals in Qaemshahr, northern Iran, than those previously reported from other areas of Iran. Therefore, efforts should be focused on public health education to reduce the risk of toxoplasmosis transmission especially in the high-risk groups in this region.

Keywords: Toxoplasmosis, *Toxoplasma gondii*, Rural health centers, ELISA, Seroepidemiologic studies

Background

Toxoplasmosis is caused by *Toxoplasma gondii*, an obligate intracellular protozoan parasite, which is commonly found in the fields as the definitive hosts, as well as a wide range of warm-blooded animals and human as the intermediate hosts (1,2). This infection is estimated to affect nearly one-third of the world's population (2-4). Humans are generally infected by the ingestion of the tissue cysts through consumption of raw or undercooked meat, or oocyst-contaminated water and foods. Vertical transmission of the parasite from mother to the fetus is regarded as a vital threat in pregnant women (5-7). Toxoplasmosis is often asymptomatic, mild, and self-limited but it can be severe and complicated in the immunocompromised individuals, such as patients with HIV/AIDS and cancer, or organ recipients (7-10). Primary infection in pregnant women can lead to

chorioretinitis, hydrocephalus, microcephaly, miscarriage, and fetal splenomegaly (6).

Seropositivity of *T. gondii* can widely vary between different areas of the world. Numerous features influence the prevalence of this infection which include nutritional habits, setting (rural or urban), socioeconomic status, and optimum moisture and temperature conditions (11,12). Seroprevalence of *T. gondii* varies from 10% to 70% in Asia (13), 16% to 80% in Europe and America (14). It seems that the seroprevalence of toxoplasmosis is around 50% in the north of Iran due to the optimum moisture and temperature conditions (10). Seropositivity of *T. gondii* has been reported to range from 22.5% to 72.5% in Mazandaran province in previous studies (15,16). However, no independent study has been done so far in Qaemshahr. Given the limited background data and lack of information on the toxoplasmosis status in

this region, the present study was designed to evaluate seroepidemiology of toxoplasmosis and risk factors of infection among the rural population in Qaemshahr, Mazandaran province, northern Iran.

Methods

Study Population

This cross-sectional study was performed from February to April 2019. Blood samples of 350 individuals referred to five laboratories of the rural health centers in Qaemshahr, Mazandaran province, Iran, were obtained through venipuncture. The age of the participants ranged between 7 and 92 years old. All the *sociodemographic* data including age, gender, job, contact with the cat or other domestic animals, dietary habits such as consumption pattern of the vegetable, drinking water, meat, milk, and egg, as well as contact with the soil were recorded through coded questionnaires after obtaining informed written consent from all the participants.

Serological Test

Sera were isolated after centrifugation at 2500 rpm for 5 minutes in the laboratory of the Razi hospital affiliated with the Mazandaran University of Medical Sciences and were kept at -20°C in properly labeled Eppendorf tubes until use. All sera were assessed for anti-*T. gondii* (IgG and IgM) antibodies, using the commercially available ELISA kits (Pishtazteb, Iran). Amounts higher than 1.1 IU/mL were considered positive and those lower than 0.9 IU/mL were considered negative according to the manufacturer's instructions. The values between 0.9 and 1.1 were considered suspicious and were tested again. Serum samples with high lipid content (hyperlipidemia) and hemolysis samples were removed in order to improve the accuracy of the test.

Statistical Analysis

Statistical analyses were performed by the SPSS software version 18.0. The relative proportions were calculated based on 95% confidence interval (CI) using chi-square and Fisher exact tests at a significant level of 5%.

Results

Among 350 sera, 296 (84.57%, 95% CI: 80.07-88.2%) were positive for anti-*T. gondii* IgG antibody (73 of 81 male (90.12%) and 223 of 269 female (82.9%) participants). The majority of seropositivity was found in female participants (76.9%, 269 cases of 350). The IgG antibody against *T. gondii* was negative in 54 (15.43%) cases. No patient was found to be positive for IgM antibody against toxoplasmosis. There was a statistically significant correlation between toxoplasmosis and age ($P=0.000$), contact with soil ($P=0.000$), and job

($P=0.005$). There was no significant association between other variables and toxoplasmosis. Table 1 shows the data of the mentioned socio-demographic characteristics.

Discussion

In the present study, the seroprevalence of anti-*Toxoplasma* IgG antibody was reported to be 84.57%, which was significant in those over 35 years of age, college students, and those having contact with the soil ($P=0.000$). In accordance with the results of the present study, Sharif et al reported that the prevalence of IgG antibody against *T. gondii* was 74.65% and Ajami et al reported a seropositivity of 86.3% in the women referred to the health centers of different parts of Mazandaran (17, 18). Our results showed that the rate of *T. gondii* infection was significantly lower in the college students than the others, which might be due to the greater access of this group to the health centers in the rural areas of Qaemshahr and the increased awareness to control their health conditions compared to other groups. Seropositivity was 90.12% in men, while it was 82.89% in women. However, no significant correlation was found, attributing to the presence of the men in jobs, such as agricultural practices and animal husbandry.

No significant association was found between the seropositivity of *T. gondii* and contact with the cats and other domestic animals. There was no association between the consumption of the unhealthy and non-chlorinated water, undercooked meat or raw egg, milk, and vegetables. A significant relationship was found between *T. gondii* infection and age higher than 35 ($P=0.000$). The seroprevalence of this protozoan parasite increased dramatically in people over 35 years of age. In fact, the level of antibodies increases with the increase in the age probably due to more exposure to *T. gondii* infection as confirmed by the previous studies (12,16,17). Additionally, in northern Iran, most people are exposed to *T. gondii* infection by the age of 15 (17).

Contact with soil was also one of the risk factors in this study. Humidity and type of soil are effective factors in the development of various parasites as well as *T. gondii* oocysts in northern Iran (19); under these optimum conditions, oocysts are easily sporulated and become prepared to establish the infection in the hosts. The prevalence rate of toxoplasmosis is between 7% and 30% in most of the South Asian countries (20); however, in Iran which is located in the southwest of Asia, it was reported to range from 9% to 86.3% (21). This great variation has been attributed to socioeconomic conditions in particular as well as diet and hygienic habits, as *T. gondii* can be transmitted through contact with cat and ingestion of contaminated meat and other foods (22). Transplacental transmission has great clinical significance and justifies the efforts aimed at

Table 1. Socio-demographic Characteristics of the Study Population for the Presence of Anti-*Toxoplasma gondii* Antibodies in a Rural Area of Qaemshahr, Northern Iran

Variables	Total No. (%)	IgG Negative No. (%)	IgG Positive No. (%)	P Value
Gender				0.119
Male	81 (23.1)	8 (9.8)	73 (90.12)	
Female	269 (76.9)	46 (17.1)	223 (82.89)	
Age (y)				0.000
<15	9 (2.57)	5 (55.5)	4 (44.44)	
15-34	75 (21.42)	27 (36)	48 (64)	
35-54	134 (38.28)	15 (11.1)	119 (88.8)	
55-74	119 (34)	6 (5.04)	113 (94.95)	
>75	13 (3.71)	1 (7.7)	12 (92.3)	
Job				0.005
Housewife	215 (61.4)	32 (14.8)	183 (85.11)	
College student	22 (6.3)	12 (54.5)	10 (45.45)	
Farmer	38 (10.9)	1 (2.6)	37 (97.36)	
Retired	17 (4.9)	1 (5.8)	16 (94.11)	
Medical staff	15 (4.2)	5 (33.3)	10 (66.6)	
Self-employed	43 (12.2)	3 (6.97)	40 (93.03)	
Rural health centers				0.896
Qadikola (1)	34 (9.7)	6 (17.6)	28 (82.5)	
Sarookola (2)	40 (11.4)	6 (15)	34 (85)	
Arateh (3)	62 (17.7)	8 (12.9)	54 (87.09)	
Alamashir (4)	82 (23.4)	11 (13.41)	71 (86.58)	
Qarakheil (5)	132 (37.7)	23 (17.42)	109 (82.57)	
Source of water				0.819
Well or watering hole	15 (4.28)	2 (13.33)	13 (86.66)	
Tap water	335 (95.81)	52 (15.5)	283 (84.47)	
Contact with domestic animals				0.059
Yes	81 (23.14)	7 (8.6)	74 (91.35)	
No	269 (76.85)	47 (17.47)	222 (82.52)	
Contact with cat				0.535
Yes	34 (9.71)	4 (11.76)	30 (88.23)	
No	316 (90.29)	50 (15.8)	266 (84.17)	
Contact with soil				0.000
Yes	15 (4.28)	2 (13.33)	13 (86.66)	
No	335 (95.81)	52 (15.52)	283 (84.47)	
Preparation and ingestion of undercooked meat, egg and raw milk				0.535
Yes	34 (9.71)	4 (11.76)	30 (88.23)	
No	316 (90.29)	50 (15.82)	266 (84.17)	
Ingestion of raw vegetables				0.301
Yes	301 (86)	44 (14.6)	257 (85.38)	
No	49 (14)	10 (20.4)	39 (79.59)	

increasing the understanding regarding the immune status of the people due to the occurrence of severe forms of toxoplasmosis in the newborn or fetus; therefore, during the pregnancy, women who might be affected with *T. gondii* infection must be cautiously observed and specific treatment should be administered to them to avoid the

placental transmission. The factors that are traditionally associated with a higher risk of toxoplasmosis were found in the rural regions of Qaemshahr. For example, in these areas, there are many stray cats in the villages and they may potentially contaminate the water, soil, and vegetables with feces. The majority of the population consumes raw

vegetables, and there is also the possibility of contact with the soil through agricultural practices. Additionally, a low level of education is more common in rural areas than in urban areas, which is associated with the increased risk of infection (23).

Under these conditions, detection of the seropositivity of IgG antibody against toxoplasmosis in 84.57% of the tested subjects at all ages was compatible with the gradual increase in the values observed in those with higher age. The results of the current project are not in consistent with several previous epidemiological studies because no significant association was found between *T. gondii* infection and contact with the cat or other animals (24,25). Variables, such as urbanization, changes in the lifestyle and habits, and better conditions of environmental sanitation and food care appear to influence the prevalence of the *T. gondii* infection (26). In addition, negative IgM antibody results in this study show that no one was in the acute phase of infection. Similar to the results of the present study, Heidari did not find any IgM antibody among high school girls in their study (20). However, Arefkhan et al reported that seropositivity for IgM antibody was 0.6 % in healthy individuals, 2.1% in hemodialysis, and 2% in cancer patients (5). El Deeb in Egypt found a seropositivity rate of 2.8% for IgM among the antenatal population (27), while, in a study conducted by Khodashenas et al, IgM seropositivity was determined to be 2% (12). Although studies on the prevalence of IgM against *T. gondii* is limited, it seems that due to the high prevalence of IgG antibodies in adults older than 35 years, the acute infection occurred at young ages in this region.

Qaemshahr is located in Mazandaran province that is a province with a mild climate accompanied by high humidity in the north of Iran (19). Probably, the high prevalence of *T. gondii* antibodies is related to the presence of stray cats in the villages, as well as high humidity, and type of soil for sporulation of this protozoan parasite.

According to the results of this study, age over 35, contact with the soil, and some occupations were the most important factors in the transmission of toxoplasmosis in this region. Therefore, due to the risk of infection with this disease for the people in this area, efforts should be focused on the public health education to reduce the risk of toxoplasmosis transmission especially in the high-risk groups. In general, the present study conducted in this rural area is one of the works indicating a high seroprevalence of this infection in Iran, which is even higher compared to the previous few studies performed in this region (16-18). This high seroprevalence of *Toxoplasma gondii* also has a positive point. Since the *T. gondii* infection during the pregnancy may cause abortion and other complications, this high prevalence of toxoplasmosis can also be helpful in the identification of premarital women of this region.

Conflict of Interests

The authors declare that there was no conflict of interests.

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Ethical Approval

The research was performed in accordance with the principles and ethical considerations of working with laboratory animals, and it was approved by the Ethics Committee of Hamadan University of Medical Sciences (Ethics Committee Code: IR.UMSHA.REC.1398.195).

Authors' Contributions

All authors have participated in conception, design, analysis, interpretation of the project, drafting the article, revising it critically for important intellectual content, and approval of the final version.

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