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Original Article

# Seroprevalence of *Toxoplasma gondii* in Women Referring to Health Centers in Sabzevar, Northeast of Iran (2021-2022)

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## Abstract

**Background:** Toxoplasmosis is a common zoonotic disease in the world, which is caused by an intracellular protozoan parasite called *Toxoplasma gondii*. In its congenital form, the pathogen is transferred to the fetus through placenta. The acquired infection is caused by either consuming contaminated meat and vegetables or contact with cats. The present research aimed to investigate the prevalence of anti-*T. gondii IgG* antibody in women referring to health centers for premarital tests in Sabzevar, Northeast of Iran (2021-2022).

**Methods:** This cross-sectional descriptive-analytical study included 190 women admitted to health centers in Sabzevar for premarital tests. The participants were selected by simple random sampling. A questionnaire was used to collect the clients' demographic information and data on some risk factors. In order to perform the tests, 2 mL of their blood samples was taken and their serum was separated. The serum samples were kept at -20 °C until the test was performed. To perform the test, after thawing the serum samples, the presence of anti-*T. gondii* IgG antibody was examined using the ELISA method in the immunology laboratory of the medical school. The data analysis was done using the chi-square test in SPSS.

**Results:** The results showed that out of 190 participants, 171 had no history of infection with this parasite and only 19 of them had a positive anti-*T. gondii IgG* antibody test result. Additionally, 90% did not have enough immunity. The Chi-square test showed a significant positive relationship between the presence of anti-*T. gondii* IgG antibody and age, education, consumption of semi-cooked meat and vegetables ($P<0.05$).

**Conclusion:** The results suggest that the positive rate of IgG test in this Northeast Iranian region is lower than in other parts, and this problem may be attributable to its specific climatic conditions, where it is hot and dry, and the humidity level is relatively low. The destruction of the parasite oocyst due to these factors can result in a low rate of parasite transmission. Therefore, the need for a screening program is not necessary, but educational programs are recommended to educate women before marriage.

**Keywords:** Seroepidemiologic studies, Immunoglobulin G, Women, Toxoplasmosis, Iran

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## Introduction

Toxoplasmosis is a common zoonotic disease in the world (1,2), caused by *Toxoplasma gondii*, an intracellular protozoan parasite that can cause severe infections in humans and animals (3). The final hosts of this parasite are cats, so that the sexual cycle of this parasite occurs only in this species of animals (4). Intermediate hosts of *T. gondii* can become infected through several routes, including ingestion of sporulated oocysts, consumption of meat of infected sheep and pigs, contact with tachyzoites, or transmission through the placenta (5). *T. gondii* infects a wide range of animals, including mammals and birds (6). It also infects cattle such as sheep and goats, causing significant economic losses that lead to abortion (7). The lambs that survive, especially in the first week of birth, can infect humans too (8). The other mammals can be infected with the parasite by swallowing food and water contaminated with oocysts shed in cat feces (9). Toxoplasmosis can be congenital or acquired. The acquired infection occurs either by ingesting oocysts excreted by an infected cat or by eating undercooked vegetables and semi-cooked meat (containing cysts) (10). The infection is usually transmitted through the consumption of water and vegetables contaminated with oocysts, use of raw and half-cooked meat contaminated with parasitic tissue cysts, placental transmission,

contaminated needles and syringes, blood and leukocyte injections, organ transplants, or even rarely by accidental injection in the laboratory (11).

About 25%-30% of the world's human population is infected with *T. gondii* parasite (12). The prevalence of Toxoplasmosis is variable among different countries and even different communities of a region of a country (13). Lower prevalence rates (10%-30%) are seen in North America, Southeast Asia, Northern Europe, and African coastal countries. Moderate rates (30%-50%) are observed in central and southern European countries, while higher prevalence rates are reported from Latin America and tropical African countries (14). In a systematic review and meta-analysis study, the prevalence of *T. gondii* infection in the general population of Iran was reported to be 39.3%. In this report, the highest and lowest rates of infection with this parasite were reported from Gilan province (86.3%) and Khuzestan province (12%), respectively (15).

Although *T. gondii* infection is asymptomatic in adults, it can threaten the lives of people, especially infants, by endangering their immune system (16). The congenital toxoplasmosis during early human pregnancy may lead to fetal death or cause retinochoroiditis, hydrocephalus, microcephaly, and jaundice in infancy (17,18). The rate of fetal transmission in the first, second, and third trimesters is 10 to 25, 30 to 40, and 60 to 65%, respectively (3). The number of children with congenital toxoplasmosis in Iran is estimated to be between 1200 and 5250 (3200 on average) annually; according to these statistics, millions of dollars are spent annually for the care and treatment of these children with congenital toxoplasmosis (19). Therefore, women's immune status is an important indicator of the occurrence of infection complications. For this purpose, in 1993, the American College of Obstetrics and Gynecology suggested serological Screening before pregnancy (20). Until now, there has been no coordinated action and diagnostic efforts to prevent the infection in pregnant women (15). Particularly, girls at the age of marriage are considered a high-risk group, because the possibility of transferring the parasite to the fetus during pregnancy rises due to their work conditions and their inadequate immunity against *T. gondii* as mothers. Considering the above-mentioned risks caused by toxoplasmosis, it seems mandatory to determine the prevalence of toxoplasmosis, closely investigate the ways of parasite transmission, and identify the high-risk groups and risk factors affecting parasite transmission in order to adopt strategies to be included in educational programs by the community health centers. Studies have already revealed that a low serum level of IgG antibody means a lack of immunity to toxoplasmosis and a higher possibility of infection in later years of life, especially during pregnancy, which can pose risks to the fetus of infected mothers. Therefore, this study was designed to determine the prevalence of anti-*T. gondii IgG* antibody in women referring to the laboratory of health centers in Sabzevar, Iran, for premarital tests for the first time. Additionally, we aimed to identify the immune status of women at marriage age in Sabzevar, Iran, regarding toxoplasmosis.

## Materials and Methods

### *Sample Size and Sampling Method*

According to a related study (20), a sample size of 200 was calculated using Cochran's formula, considering $P = 0.4$, $d = 0.05$, and $\alpha = 0.05$. However, the actual sample size was 190 persons.

$$N = \frac{z^2\left(1-\frac{\alpha}{2}\right)\times P(1-P)}{d^2} = 3.84\times 0.19\times 0.81 \div 0.00295483 = 200$$

This descriptive-analytical cross-sectional study was conducted including 190 women at the age of marriage referring to the medical laboratory of health centers in Sabzevar for pre-marriage tests using the random sampling method based on their accessibility and availability to the researcher.

A questionnaire containing the clients' demographic information and relevant risk factors, such as eating habits and having a cat, was designed and completed through face-to-face interviews. Moreover, to perform the medical tests, 2 mL of their blood samples was taken from each participant, and the serum was then separated. Serum samples were kept at -20 °C until performing the tests. After thawing the serum samples, the presence of IgG anti-*T. gondii* antibody was determined using the ELISA method according to the manufacturer's instructions (Danish Banyan Company) in the immunology laboratory of the medical school.

### *Statistical Analysis*

The SPSS software version 26.0 was used for data analysis. Descriptive statistics including frequency, percentage, mean ± standard deviation, and variance, as well as inferential statistical methods (i.e., the chi-square test), were used in this research.

## Results

The results showed that the mean age of the participants was 27.152 ± 9.024 years. The minimum and maximum ages of study participants were 14 and 57 years, respectively.

According to the results presented in Table 1, out of 190 women studied, 50 (26.3%) had an education level below a high school diploma, 67 (35.3%) had a high school diploma, and 20 (10.5%) had an associate degree. In addition, 36 (18.9%) had a bachelor's degree and 17 (8.9%) had graduate and postgraduate degrees.

### *Determining the Immunity of Women Against Toxoplasmosis Across Different Age Groups*

As Table 1 shows, 91 women (96.8%) under 24 years old and 80 women (83.4%) over 24 years old were negative for anti-*T. gondii IgG* antibody test. Additionally, 3 (3.2%) subjects were under 24 years old and 16 women (16.6%)

**Table 1.** Epidemiological Characteristics of Investigated Women

| Variable | | Negative IgG | | Positive IgG | | *P* Value |
|---|---|---|---|---|---|---|
| | | No. | % | No. | % | |
| Age | ≤24 | 91 | 96.8 | 3 | 3.2 | 0.002 |
| | <24 | 80 | 83.4 | 16 | 16.6 | |
| Education | Below high school diploma | 41 | 21.58 | 9 | 4.7 | 0.024 |
| | High school diploma | 60 | 31.58 | 7 | 3.68 | |
| | Associate degree | 19 | 10 | 1 | 0.526 | |
| | Bachelor's degree | 36 | 18.9 | 0 | 0 | |
| | Graduate and postgraduate | 15 | 7.89 | 2 | 1.05 | |
| Consumption of raw meat | No | 171 | 90 | 18 | 9.47 | 0.003 |
| | Yes | 0 | 0 | 1 | 0.526 | |
| Consumption of undercooked vegetables | No | 19 | 10 | 0 | 0 | 0.046 |
| | Yes | 152 | 80 | 19 | 10 | |
| Contact with cat | No | 150 | 78.9 | 19 | 10 | 0.105 |
| | Yes | 21 | 11.1 | 0 | 0 | |

were over 24 years old. The Chi-square test showed that there was a significant relationship between the presence of anti-*T. gondii IgG* antibody and age ($P=0.002$) in women. This means that the prevalence of the anti-*T. gondii IgG* antibody test was higher in older women.

### *Determining the Immunity of Women Against Toxoplasmosis in Patients with Different Literacy Levels*

As Table 1 shows, most of the participants who had a positive anti-*T. gondii IgG* antibody test had a bachelor's degree (n=9). The results of the chi-square test showed that there was a significant relationship between immunity to toxoplasmosis and level of education ($P=0.024$) in women. This means that the prevalence of anti-*T. gondii IgG* antibody was higher in women with lower education levels.

### *Determining the Immune Status of Women Across Different Groups in Terms of Consumption of Under-Cooked Vegetables, Semi-cooked Meat, and Contact With Cats*

As Table 1 shows, out of 19 women whose IgG anti-toxoplasma antibody tests were positive, only one participant had consumed half-cooked meat. In other words, 18 cases whose test results were positive had a risk factor other than the consumption of undercooked vegetables or half-cooked meat, which had a positive effect on the test result. Additionally, 100 % of 19 women with positive IgG anti-*T. gondii* antibody test consumed vegetables. The chi-square test also showed that there was no significant relationship between immunity to toxoplasmosis and contact with cats ($P=0.105$) in women. However, there was a significant positive relationship between immunity to toxoplasmosis and consumption of vegetables and consumption of semi-cooked meat ($P=0.003$).

## Discussion

The results of the present study showed that out of 190 women who were about to get married, 19 (10%) had anti-toxoplasma antibody (IgG), which is lower than the prevalence rate in other parts of Iran. This issue may be attributable to the specific weather conditions, where the climate is typically hot and dry, and the humidity level is relatively low, which causes the oocysts of the parasite to deteriorate, causing a decrease in the rate of parasite transmission. On the other hand, the low prevalence rate means that the population is still susceptible to the disease, predisposing this sensitive population to the risk.

It was shown that there was a significant relationship between the presence of anti-*T. gondii IgG* antibody and age in women; therefore, as age increased, the positive rate of IgG antibody test against *T. gondii* increased too. This is because as the age increases, exposure to pathogenic factors increases and the probability of disease also increases.

However, this relationship was reported to be statistically significant across different groups in terms of education level. In other words, the rate of positivity of IgG antibody test against toxoplasmosis increased at lower education levels, because at higher education levels and advanced literacy, the level of awareness of individuals also increased, which led to decreased levels of infection and the incidence of the disease.

The results also showed that there was no significant relationship between immunity to toxoplasmosis and contact with cats in women. However, there was a positive and statistically significant relationship between immunity to toxoplasmosis and the consumption of semi-cooked meat, as well as the consumption of under-cooked vegetables.

Jalali et al conducted a study to determine the seroepidemiology of toxoplasmosis in pregnant women referring to the laboratories of health centers in Bojnord, Iran. A total of 211 pregnant women were included in their study. Based on the results, 30.8% of women participating in their study had IgG antibody against

*T. gondii*. Their findings showed that contact with a cat increases the chances of contracting this infection. However, no significant relationship was observed between the consumption of raw or undercooked meat, the way of washing, and the consumption of unwashed vegetables and the positive rate of antibodies (21).

Fallah et al designed a study with the aim of determining the prevalence of toxoplasmosis in primiparous women in Hamadan, Iran, and included 576 participants, whose data were analyzed using IFA method for total antibody against *T. gondii* in their serum using a questionnaire and blood test (22). The prevalence rate of toxoplasmosis was 33.5%, and the results showed that there was a statistically significant relationship between the prevalence of contamination and age, the way of meat consumption, the use of fresh meat in the preparation of kabab, and the consumption of raw vegetables. On the other hand, no significant relationship was observed between the prevalence of contamination and education, contact with cats and raw meat, the method of washing vegetables, and consumption of raw milk and half-boiled eggs (22).

Mahmoudi et al conducted a study titled "Seroepidemiological study on toxoplasmic infection among high-school girls by IFAT in Esfahan city, Iran", and analyzed the data obtained from 414 high school girls from different areas of Isfahan city. Their findings showed that among the variables they examined, there was only a significant relationship between the history of contact with cats and the history of keeping birds at home and the amount of anti-*T. gondii* immunoglobulin; however, no significant relationship was found between other factors such as people's level of awareness, eating habits, occupation of parents, and so on. No symptoms such as fever, lymphadenopathy, and so on were seen in any of the subjects. Based on the results, it can be seen that more than 80% of the people studied were serologically negative using the Indirect Fluorescent Antibody Test (IFAT), and according to the sensitivity of these people, there is a possibility that the mothers and their children will be infected with toxoplasmosis in the future (23).

In another study titled "Seroepidemiology of toxoplasmosis in pregnant women referring to health centers in the city of Hamadan in 1391", 350 pregnant women admitted to the health centers of Hamedan city were included in the study. In their study, written informed consents were obtained from the volunteers, and their serum samples were tested by IgG ELISA and IgM ELISA methods (24). Their findings showed that due to the significant relationship between the disease and pregnant women's age, occupation, and level of education, necessary training and awareness should be provided for the prevention and care of toxoplasmosis (24).

In 2012, Heydari et al conducted a study with the aim of determining the prevalence of IgM and IgG antibodies against *T. gondii* and predisposing factors for infection among 420 high school girls in Gonabad, Iran. Their findings showed a statistically significant relationship between the consumption of semi-cooked meat and raw vegetables and the positivity of anti-*T. gondii* IgG (25).

In 2017, in another study titled "Seroepidemiological study of toxoplasmosis in females for pre-marriage evaluation", Rezaei et al investigated 400 women who were referred for pre-marriage tests and found that there was a significant relationship between infection with this parasite and age. However, no statistically significant relationship was observed between pollution and place of residence, contact with cats and other pets, consumption of grilled and semi-cooked meat, consumption of raw milk, use of gloves when slicing meat into pieces, and ways of washing vegetables (26).

In the study conducted by van den Berg et al in the Netherlands, the prevalence of IgG antibody against *T. gondii* was investigated in women aged 15-45 years, and the average seroprevalence in this group of women was reported to be 17.5% (27).

In another study by Wodage et al, 208 serum samples were obtained from pregnant women in Merket, Northeast of Ethiopia, and were examined by latex agglutination test, where 32.2% had a positive test result (28).

In a study conducted by Daka et al in Namwala, Zambia (2021), 401 serum samples from pregnant women aged 24-30 were examined, and it was found that 2.4% were positive for anti-*T. gondii* antibodies (29).

In another study conducted by Mustafa et al in Zakho, Iraq, 610 women aged 18-79 were examined, and 32.46% of them were found to be positive for anti-*T. gondii* antibodies (30).

In a survey conducted by Shabanian et al in 2023 in Ramsar, North of Iran, 191 pregnant women were examined by ELISA method for the presence of anti-*T. gondii* IgG, and it was found that 46.1% were positive (31).

After reviewing the studies of other researchers and comparing them with the present study conducted in Sabzevar, Northeast of Iran, we came to the conclusion that according to the weather conditions of each region of the country and based on the difference in the amount of heat and humidity, the rate of infection and the number of positive tests will be different in terms of anti-*T. gondii IgG* antibody titer. In other words, the more the climatic heat increases and the humidity decreases (as in Sabzevar, Iran), the lower the frequency of infection will be, suggesting that the duration of the disease-causing agents in hot and dry weather conditions will be shorter in comparison with areas which have sub-humid and humid weather conditions.

## Conclusion

The results of the present study showed that the positive rate of IgG test is lower in Sabzevar, Northeast of Iran, than in many parts of the country, and this difference is attributable to its hot and dry weather conditions, where the humidity level is relatively low. Consequently, the parasite oocyst is destroyed, which can cause a decrease in the rate of parasite transmission. Therefore, the need

for a screening program is not felt, but it would be better if an educational program were developed for women at the age of marriage.


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## Authors' Contribution

**Conceptualization:** Hossein Elyasi.
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**Formal analysis:** Anis Baghi keshtan.
**Funding acquisition:** Hossein Elyasi.
**Investigation:** Anis Baghi keshtan.
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**Validation:** Hossein Elyasi.
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**Writing–original draft:** Hossein Elyasi.
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## Competing Interests

None declare.

## Ethical Approval

This study was approved by the Ethics Committee of Sabzevar University of Medical Sciences (IR.MEDSAB.REC.1399.162).


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