



# Seroprevalence of Specific Antibodies Against *Toxoplasma gondii* Among Blood Donors in Abadan and Khorramshahr Cities, Southwest Iran

Masoud Foroutan <sup>1</sup>, Shahrzad Soltani <sup>1,\*</sup>, Haniyeh Badpa <sup>2</sup>, Fatemeh Maghsoudi <sup>1</sup>, Mohammad Amin Shaverdi <sup>1</sup>

<sup>1</sup> Department of Basic Medical Sciences, Faculty of Medicine, Abadan University of Medical Sciences, Abadan, Iran

<sup>2</sup> Student Research Committee, Abadan University of Medical Sciences, Abadan, Iran

\*Corresponding Author: Department of Basic Medical Sciences, Faculty of Medicine, Abadan University of Medical Sciences, Abadan, Iran. Email: shahrzadsoltani225@yahoo.com

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## Abstract

**Background:** *Toxoplasma gondii* has emerged as a concern regarding the potential transmission from blood donors to recipients.

**Objectives:** Several studies have been published on the epidemiology of *T. gondii* in blood donors; however, the present study was conducted to determine the seroprevalence of *T. gondii* in blood donors in the cities of Khorramshahr and Abadan, Southwest Iran.

**Methods:** In this cross-sectional study, data were collected via direct sampling in 2022. The presence and levels of *T. gondii*-specific IgG and IgM antibodies in blood donors were investigated using enzyme-linked immunosorbent assay (ELISA). A logistic regression model was employed to examine the association between *T. gondii* seropositivity and demographic variables and risk factors.

**Results:** A total of 345 blood donors were included in this study, of whom 110 (31.88%) had IgG seropositivity, while IgM seropositivity was detected in 6 donors (1.7%). Both IgG and IgM seropositivity were found in 5 donors (1.5%). Statistical analysis revealed no significant associations between *T. gondii* seropositivity and the investigated demographic and behavioral variables.

**Conclusions:** This study provides valuable insights into the seroprevalence of *T. gondii* in blood donors in Khorramshahr and Abadan, as well as socio-demographic and behavioral risk factors, serving as a basis for future prevention programs.

**Keywords:** *Toxoplasma gondii*, Enzyme-Linked Immunosorbent Assay, Seroprevalence, Iran

## 1. Background

*Toxoplasma gondii* is considered one of the most prevalent parasitic infections worldwide, with approximately one-third of the global population being seropositive for the parasite. The infection has a global seroprevalence, with higher prevalence rates in tropical and subtropical areas (1, 2). Infection typically occurs through the ingestion of food or drinking water contaminated with oocysts, consumption of raw or undercooked meat containing tissue cysts, vertical transmission from mother to fetus, and less commonly, through blood transfusions or organ transplantation (3-

6). The infection is typically asymptomatic in immunocompetent individuals, whereas in immunocompromised individuals, it can cause severe, progressive, and life-threatening complications (7). Patients with conditions such as aplastic anemia, sickle cell anemia, and thalassemia, who require frequent blood transfusions to survive, may experience irreparable complications from opportunistic infections such as *T. gondii* due to their immunodeficiency (5, 8). Recent evidence suggests that *T. gondii* can be transmitted via transfusion of whole blood and white blood cells, with the parasite capable of surviving for up to 50 days in blood stored at 5°C (9, 10).

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Asymptomatic blood donors in the acute phase of infection with parasitemia may play an important role in the transmission of this parasite (11-13). In Iran, the seroprevalence of *T. gondii* varies considerably across different geographical regions (14). In recent decades, transfusion-transmitted infections (TTIs), including *T. gondii* from blood donors, have become a global concern for blood recipients, particularly among immunocompromised populations (5).

## 2. Objectives

It is worth mentioning that screening for *T. gondii* before transfusion has not been widely implemented. Since there is no evidence regarding the epidemiological data showing the seroprevalence of this infection in donated blood in this region in past years, the present research aimed to examine the seroprevalence of IgG and IgM antibodies against *T. gondii* among apparently healthy blood donors in the cities of Abadan and Khorramshahr, located in Khuzestan province, Iran.

## 3. Methods

### 3.1. Study Area

Abadan and Khorramshahr are cities located in Khuzestan province in southwestern Iran. Both cities are situated on flat plains and experience a hot desert climate. There is a noticeable variation in air temperature between summer and winter, with summer temperatures exceeding 50°C and winter temperatures typically ranging from 16°C to 20°C. Annual dust storms occur in this area. The average annual humidity is 45%, and the region is generally considered humid, with humidity levels reaching 100%.

### 3.2. Population

This cross-sectional study included 345 healthy volunteer blood donors who referred to central Blood Transfusion Organizations (BTO) affiliated with Abadan University of Medical Sciences (AUMS) in Abadan and Khorramshahr between May and September 2022. Convenience sampling was used due to logistical constraints; future studies should prioritize powered calculations. Written informed consent was obtained and signed by all participants prior to enrollment. Blood donations were routinely screened for five pathogens, including human T-lymphotropic virus 1 and 2 (HTLV1, 2), Hepatitis C virus (HCV), Hepatitis B virus (HBV), human immunodeficiency virus (HIV), and *Treponema pallidum*, in accordance with BTO guidelines (15). The

study followed three inclusion criteria to select participants: (1) Volunteers aged  $\geq 18$  years; (2) individuals who provided informed consent to participate in the investigation; and (3) donors whose blood tests were negative for HTLV1, HTLV2, HCV, HBsAg, HIV, and *T. pallidum*. A structured questionnaire was completed by each blood donor, consisting of demographic information and several risk factors associated with *T. gondii* infection.

### 3.3. Serology

Approximately 10 mL of venous blood was collected from each eligible participant. After collection, the blood samples were centrifuged at 3500 rpm for 5 minutes in anticoagulant-free tubes. The serum samples were then separated and stored at -20°C until further analysis. The serum samples were evaluated for specific IgG and IgM antibodies against *T. gondii* using Torch-IgG, IgM-Trinity (Biotech Company), following the manufacturer's protocols. Samples with values less than 0.9, between 0.9 and 1.1, and above 1.1 were considered negative, borderline, and positive, respectively (16). Sensitivity and specificity for IgG were 90 - 100% and 95 - 100%, respectively. For IgM, these values were 80 - 100% and 80 - 100%, respectively. The results were recorded by an enzyme-linked immunosorbent assay (ELISA) reader based on optical density (OD) values, and the antibody concentrations were interpreted according to the OD thresholds provided by the manufacturer.

### 3.4. Statistical Analysis

The data were analyzed descriptively following the collection of laboratory results and questionnaire responses. Descriptive statistics, including frequency and percentage, were used. The chi-square test was applied using SPSS software version 19 (SPSS Inc., Chicago, IL, USA). Furthermore, univariate logistic regression analysis was performed to evaluate the probable association between the seroprevalence of *T. gondii* infection (based on IgG antibody) and related risk factors. The level of significance was set at  $P < 0.05$ .

## 4. Results

In this study, 97.1% of the blood donors were male. Among the participants, 31.9% were IgG positive, and 1.5% were positive for both IgM and IgG antibodies (Table 1). The majority of participants (42.02%) were aged between 36 and 45 years, and 91.9% lived in residential areas located in cities. Of the 345 participants, 6 individuals (1.8%) tested positive for IgM antibodies. Logistic regression analysis showed no significant association

between the variables studied. Notably, 95.7% of participants reported not consuming raw or semi-cooked meat. Despite this, 31.5% of these individuals were IgG positive, and 1.5% were positive for both IgM and IgG antibodies; however, statistical analysis did not reveal a significant relationship between these cases (Table 2). Additionally, 78.3% of participants consumed properly washed vegetables, among whom 30.7% were IgG positive, with no significant association observed. Regarding contact with cats, 95.9% of cases had no contact with cats, yet 31.7% and 1.5% of these individuals were positive for IgG and both IgG/IgM antibodies, respectively. Among those who had contact with cats, no meaningful association was noticed with IgG positivity. Furthermore, 15.1% of participants had contact with soil, among whom 21 individuals (36.9%) were IgG positive. However, statistical analysis did not reveal any significant relationship. Among the participants, 115 individuals (33.3%) had blood type O positive, and 29.6% of these individuals were IgG positive. No significant relationship was observed between IgG positivity and blood type O positive. Despite statistical non-significance, the 42.9% IgG seroprevalence in rural donors (vs. 30.9% in urban donors) warrants further study due to potential environmental exposure differences.

## 5. Discussion

Given the necessity of reducing toxoplasmosis transmission and mitigating the complications of this infection, this cross-sectional study investigated the seroprevalence of *T. gondii* IgG and IgM antibodies among blood donors in Khorramshahr and Abadan, Iran, and examined associated epidemiological factors. Demographic characteristics and feeding behaviors were assessed alongside blood sampling for *T. gondii*. The main findings showed that the seroprevalence of *T. gondii* IgG antibodies among participants was 31.88% (110/345). This seroprevalence was higher than in previous studies conducted in regions such as Turkey (17), Mexico (18, 19), Chile (20), India (12), Malaysia (21), Mali (22), and Thailand (23), but aligns with global averages (5) and national estimates (14). Differences in *T. gondii* seroprevalence between studies may be attributed to geographic factors (5, 14). In a study aimed at evaluating the seroprevalence of infection in blood donors in China using a meta-analysis, 40 eligible studies were reviewed, reporting that among 49,784 blood donors in China, the prevalence of toxoplasmosis was 6.26% (24). The seroprevalence of *T. gondii* IgG antibody in Khorramshahr and Abadan was comparable to the global average (5). Statistical analysis of risk

factors showed that the seroprevalence of *T. gondii* was not statistically different between men and women ( $P = 0.821$ ), consistent with prior studies (25). Although blood donors with lower education levels exhibited a higher prevalence of toxoplasmosis, this association was not statistically significant. These findings could be attributed to differences in living conditions and increased likelihood of exposure to potential infection sources. Health education and promotion, especially avoiding eating raw and undercooked meat and avoiding contact with feline feces, are key recommendations to reduce the prevalence of toxoplasmosis (26). Consistent with other studies, the majority of participants in this study did not consume raw or semi-cooked meat, and no significant association was observed between this factor and *T. gondii* seropositivity (27). However, a study investigating *T. gondii* seroprevalence in blood donors in Boyar Ahmad city in Iran demonstrated that risk factors such as contact with soil and consumption of semi-cooked meat were significantly associated with *T. gondii* seropositivity (28). In fact, the consumption of semi-cooked meat, particularly lamb and goat meat containing tissue cysts, was known as an important source of *T. gondii* infection in that city (29, 30). Cultural dietary practices (e.g., thorough meat cooking) or underreported cat exposures may explain discrepancies. A hypothesis suggests that in Khorramshahr and Abadan, cities near the Persian Gulf, there is a dietary balance between the consumption of animal products and seafood. In contrast, in rural areas in northern Iran, the consumption of semi-cooked meat and unwashed vegetables are major risk factors for toxoplasmosis. Similarly, contact with cats and consuming raw vegetables and raw eggs have been recognized as risk factors for toxoplasmosis among blood donors in the southeast of Iran (30, 31). Among participants, 78.3% consumed properly washed vegetables, with 30.7% of this group testing IgG positive. However, no significant association was observed between vegetable consumption and *T. gondii* seropositivity. In a similar study, serological investigation and determination of the genotype of *T. gondii* among Iranian blood donors in Mazandaran province were conducted, and there was an association between the prevalence of *T. gondii* and sex, blood group, Rh, source of water, and meat of nymphs and contact with garden soil, contact with animals, but no significant relationship was found between drinking raw milk and consuming raw vegetables (32). This aligns with findings from other studies, which also did not identify a relationship between the consumption of vegetables and *T. gondii* infection (17, 33). Among the cases, 95.9% had no contact with cats. Among

**Table 1.** Demographic Characteristics and *Toxoplasma gondii* Seroprevalence Among Healthy Blood Donors in Southwest Iran

Variables	No. (%)	IgG Positive (%)	IgM Positive (%)	Both IgG/IgM Positive (%)
<b>Gender</b>				
Male	335 (97.1)	107 (31.9)	6 (1.8)	5 (1.5)
Female	10 (2.9)	3 (30)	-	-
<b>Age groups (y)</b>				
< 25	12 (3.47)	3 (25)	-	1 (8.3)
26 - 35	121 (35.07)	39 (32.2)	1 (1)	1 (0.8)
36 - 45	145 (42.02)	48 (33.1)	5 (3.1)	2 (1.4)
> 45	67 (19.42)	20 (29.9)	-	1 (1.5)
<b>Residence area</b>				
Urban	317 (91.9)	98 (30.9)	6 (1.9)	5 (1.6)
Rural	28 (8.31)	12 (42.9)	-	-
<b>Education level</b>				
Diploma or lower	237 (68.7)	81 (34.2)	4 (1.7)	4 (1.7)
University degree	108 (31.3)	29 (26.9)	2 (1.9)	1 (0.9)
<b>Raw/half-cooked meat consumption</b>				
Yes	15 (4.3)	6 (40)	-	-
No	330 (95.7)	104 (31.5)	6 (1.8)	5 (1.5)
<b>Eating properly washed vegetables</b>				
Yes	270 (78.3)	83 (30.7)	6 (2.2)	3 (1.1)
No	75 (21.7)	27 (36)	-	2 (2.7)
<b>Contact with cat</b>				
Yes	14 (4.1)	5 (35.7)	-	-
No	331 (95.9)	105 (31.7)	6 (1.8)	5 (1.5)
<b>Contact with soil</b>				
Yes	52 (15.1)	21 (39.6)	1 (1.9)	-
No	293 (84.9)	89 (30.5)	5 (1.7)	5 (1.7)
<b>Blood group and Rh</b>				
A (Rh positive)	90 (26.1)	36 (40)	1 (1.1)	2 (2.2)
A (Rh negative)	5 (1.4)	3 (60)	-	-
B (Rh positive)	85 (24.6)	21 (24.7)	2 (2.4)	1 (1.2)
B (Rh negative)	5 (1.4)	1 (20)	-	-
AB (Rh positive)	29 (8.4)	9 (31)	-	1 (3.4)
AB (Rh negative)	2 (0.6)	1 (50)	-	-
O (Rh positive)	115 (33.3)	34 (29.6)	3 (2.6)	1 (0.9)
O (Rh negative)	14 (4.1)	5 (35.7)	-	-

participants who had contact with cats (transient exposure), no meaningful association was observed between cat exposure and IgG seropositivity. Despite the fact that a large number of cases were not in contact with felines, other studies considered this variable as one of the main risk factors for *T. gondii* infection (34, 35). Cats are an important source of infection and can excrete large numbers of oocysts that can survive for months in humid environments. In addition, beetles and flies can spread oocysts from soil to food. Due to the amount of oocysts in the habitat, animals can be infected, which can cause an increase in infection among animals. Additionally, tissue cysts of infected

rats may transmit to cats through hunting. Cats can pollute water, gardens, fields, and food by excreting large amounts of oocysts (32). Underreported cat exposures may explain discrepancies. In the current study, 15.1% of participants had contact with soil, with 36.9% of these individuals testing IgG positive. However, statistical analysis did not reveal a significant relationship between soil contact and seropositivity. A similar study aimed at serological investigation and determining the genotype of *T. gondii* in blood donors in Iran concluded that there was no meaningful association between the seroprevalence of *T. gondii* and garden soil (32). Among the participants, 33.3% had an O-

**Table 2.** Univariate (Crude OR) Logistic Regression Analysis of the Potential Risk Factors Associated with *Toxoplasma gondii* IgG Seroprevalence Among Healthy Blood Donors of Southwest Iran

Variables	IgG Positive	
	Crude OR (95% CI)	P-Value
<b>Gender</b>		
Male	1.172 (0.297 - 4.618)	0.821
Female	1	-
<b>Age groups (y)</b>		
< 25	1.095 (0.297 - 4.045)	0.891
26 - 35	1.082 (0.57 - 2.052)	0.81
36 - 45	1.153 (0.621 - 2.142)	0.653
> 45	1	-
<b>Residence area</b>		
Urban	0.642 (0.293 - 1.406)	0.268
Rural	1	-
<b>Education level</b>		
Diploma or lower	1.454 (0.884 - 2.392)	0.141
University degree	1	-
<b>Raw/half-cooked meat consumption</b>		
Yes	1.352 (0.469 - 3.894)	0.577
No	1	-
<b>Eating properly washed vegetables</b>		
Yes	0.741 (0.436 - 1.26)	0.269
No	1	-
<b>Contact with cat</b>		
Yes	1.116 (0.365 - 3.41)	0.847
No	1	-
<b>Contact with soil</b>		
Yes	1.382 (0.757 - 2.526)	0.292
No	1	-
<b>Blood group and Rh</b>		
A (Rh negative)	3.429 (0.548 - 21.432)	0.188
A (Rh positive)	1.67 (0.938 - 2.974)	0.081
B (Rh negative)	0.571 (0.062 - 5.298)	0.622
B (Rh positive)	0.798 (0.426 - 1.494)	0.481
AB (Rh negative)	2.286 (0.139 - 37.592)	0.563
AB (Rh positive)	1.203 (0.508 - 2.85)	0.675
O (Rh negative)	1.27 (0.397 - 4.063)	0.687
O (Rh positive)	1	-

positive blood group, with 29.6% testing IgG positive. Statistical analysis did not show a significant relationship between IgG positivity and the O-positive blood group. Similar studies have confirmed that there is no association between *T. gondii* IgG prevalence and ABO or Rhesus blood groups (36, 37). These results show that blood group is not a risk factor for infection (38, 39). It seems that the ABO/Rh non-association with *T. gondii* infection (Table 2) aligns with global literature, reinforcing blood groups' irrelevance to *T. gondii* susceptibility.

### 5.1. Conclusions

The seroprevalence of *T. gondii* antibodies among blood donors in Khorramshahr and Abadan was estimated to be 31.88%. Although seroprevalence does not indicate active *T. gondii* infection, there remains a risk of toxoplasmosis transmission via blood transfusions. Given that most blood recipients are at-risk patients, it is crucial to incorporate screening for *T. gondii* antibodies into the routine protocols of Iran's



Blood Transfusion Organization, particularly in the cities of Khorramshahr and Abadan.

## 5.2. Limitations

In this study, only the ELISA serologic test was performed, which has high sensitivity (90 - 100% for IgG and 80 - 100% for IgM) and specificity (95 - 100% for IgG and 80 - 100% for IgM). The absence of molecular tests to confirm serological results, which could have enhanced diagnostic accuracy, is the main limitation of this research project.

## Footnotes

**Authors' Contribution:** S. S. and M. F. designed the study protocol. S. S., H. B., F. M., and M. A. S. collected the data and involved in statistical analysis. S. S. and H. B. performed the experiments. H. B. drafted the manuscript. S. S. and M. F. critically revised the manuscript. All authors read and approved the final version of the manuscript.

**Conflict of Interests Statement:** The authors declare no conflict of interest.

**Data Availability:** The data used to support the findings of this study are available from the corresponding author upon reasonable request.

**Ethical Approval:** The study protocol was received the approval from the Ethics Committee of Abadan University of Medical Sciences (IR.ABADANUMS.REC.1400.005).

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