Seroepidemiological evaluation of *Toxoplasma gondii* immunity among the general population in southwest of Iran

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Abstract Toxoplasmosis is a cosmopolitan zoonotic infection, caused by a unicellular protozoan parasite known as *Toxoplasma gondii* (*T. gondii*) that belongs to the phylum Apicomplexa. The present investigation was aimed to evaluate the seroprevalence of *T. gondii* infection in the general population of Abadan city. In this cross-sectional study, a total of 496 subjects were participated. Anti-*T. gondii* IgG and IgM antibodies were tested using commercially available enzyme-linked immunosorbent assay (ELISA). Moreover, a structured questionnaire was completed for each person. Out of 496 subjects, 188 (37.9%) and 30 (6.05%) samples were seropositive for IgG and IgM, respectively. The more seroprevalence was found during spring season, among female subjects, in rural inhabitants, in persons with the education level of diploma or lower, from the subjects with a history of contact with cat, in individuals who consumed raw/undercooked meat,

and amongst who drink unpurified water. The results showed that inhabitants of tropical areas, may be moderately exposed to *T. gondii*. Increase of knowledge of people about toxoplasmosis, certainly affects in reduction of the infection.

Keywords *Toxoplasma gondii* · Seroprevalence · ELISA · Abadan · Iran

Introduction

Toxoplasmosis is a cosmopolitan zoonotic infection, caused by a unicellular protozoan parasite known as *Toxoplasma gondii* (*T. gondii*) that belongs to the phylum Apicomplexa (Dubey 2008). In general, *Toxoplasma* needs two hosts to complete its life cycle in the environment. For this purpose, cats (Family: Felidae) are the definitive hosts, while the majority of warm-blooded animals serve as intermediate hosts (Dubey 2008; Foroutan et al. 2018a; Khademvatan et al. 2017).

The infection is predominantly transmitted through the following routes: consumption of raw/undercooked meat contaminated with tissue cysts, drinking water or ingestion of raw/unwashed vegetables contaminated by mature oocysts, and vertically from mother to the fetus (Dubey 2008; Foroutan-Rad et al. 2016a; Foroutan et al. 2018a). However, transmission through blood transfusion and organ transplantation are less frequent (Foroutan-Rad et al. 2016b; Foroutan and Majidiani 2018; Yousefi et al. 2017). In humans, the infection is often subclinical and asymptomatic in immunocompetent persons, although in immunocompromised subjects such as patients with malignancies, HIV-positive individuals, and organ transplant recipients may be causes severe and progressive

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complications with poor prognosis, including brain abscess, encephalitis, chorioretinitis, myocarditis, etc. or even may result in death if not treated (Fallahi et al. 2017; Foroutan et al. 2018a; Majidiani et al. 2016; Rostami et al. 2018; Wang et al. 2017). Moreover, *T. gondii* infection may cause severe complications for the fetus in those seronegative pregnant mothers that acquired the infection during their pregnancy period (Fallahi et al. 2018; Foroutan-Rad et al. 2016a). Despite of continuous attempts of scientists and great advances during three past decades, there is no commercial licensed vaccine against toxoplasmosis for medical application in humans (Foroutan and Ghaffarifar 2018; Foroutan et al. 2018b, d).

Based on recent systematic review papers with meta-analysis approach in Iran, the seroprevalence of toxoplasmosis were estimated among different human groups. The pooled exposure to *T. gondii* was relatively high in various populations including: cancer patients 45% (95% CI = 39–51), HIV/AIDS subjects 50% (95% CI = 37–63), transplant recipients 55% (95% CI = 43–67), general population 39% (95% CI = 33–46), pregnant women 41% (95% CI = 36–45), apparently healthy blood donors 33% (95% CI = 24–42), and hemodialysis patients 58% (95% CI = 46–70) (Daryani et al. 2014; Foroutan et al. 2018a, c).

Numerous published papers have determined the seroprevalence of toxoplasmosis in different human groups of Khuzestan province, southwest of Iran, indicating the people of this area has relatively moderate to high exposure to *T. gondii*. The majority of previous investigations have been performed in high risk populations, including pregnant women, immunosuppressed individuals, premarital women, hemodialysis patients, diabetic subjects, and multi-transfused thalassemia patients (Ghasemian et al. 2007; Maraghi et al. 2013a, b; Saki et al. 2013, 2015, 2016, 2017; Soltani et al. 2013; Yad Yad et al. 2014; Yousefi et al. 2017). Since there is a lack of an epidemiological data showing the prevalence of toxoplasmosis in human general population during recent years, thus, the current study was aimed to determine the seroprevalence of anti-*Toxoplasma* IgG and IgM antibodies in those subjects who referred to the Taleghani hospital, affiliated to Abadan School of Medical Sciences, southwest of Iran.

Materials and methods

Study area

Abadan city is located in the southwest of Iran (Khuzestan province) (Fig. 1). Abadan is confined in the East by the Bahmanshir outlet of the Karun River (the Arvand Rood)

and in the West by the Arvand waterway, near the Iraq-Iran border. Its population today has reached about 350,000 people. Weather temperature is highly varied throughout the year, ranged from 53 °C in summer to − 4 °C in winter. This city has a mean temperature of 25.5 °C, average relative humidity of 45%, and mean annual precipitation of 153.3 mm (https://en.wikipedia.org/wiki/Abadan,_Iran).

Study population

In this cross-sectional study, venous blood samples were taken from 496 participants who were referred to Taleghani hospital affiliated to Abadan School of Medical Sciences between July 2017 and June 2018 for routine checkups. A written informed consent was obtained from adult persons and parent or guardian of children less than 15 years old. This study received the approval from the Abadan School of Medical Sciences Ethical Committee (1396.239).

Serology

From each person who agreed to participate in this study, 5 mL of venous blood was collected. All blood specimens were transferred to the central laboratory of the Abadan School of Medical sciences. Firstly, the blood samples were centrifuged at 4000 rpm for 5 min. Then the sera were separated and stored in − 20 °C until examined. The specific anti-*T. gondii* IgM and IgG antibodies were tested by commercial available enzyme-linked immunosorbent assay (ELISA) kit (Torch-IgG, IgM-Trinity Biotech Company) based on the manufacturer's instruction. The samples with international units (IU)/mL of < 0.9, 0.9–1.1, and > 1.1 were considered as negative, borderline, and positive, respectively.

Questionnaire

A questionnaire was completed for each individual containing some demographic information, including gender (Male or Female), age (10–25, 25–40, 40–55, and 55–72), place of residence (Urban or Rural), season (Spring, Summer, Autumn, and Winter), education level (Diploma or lower and University degree), contact with cat (Yes or No), consumption of raw/undercooked meat (Yes or No), and source of drinking water (Purified or Unpurified water).

Statistical analysis

Data analysis was performed using SPSS version 22 and Chi square test.

Fig. 1 Location of Abadan city. The study region is shown with black asterisk

Results

Participants

In the current study, 496 individuals were participated from July 2017 to June 2018. Of these, 243 (49%) and 253 (51%) were male and female, respectively. The age range of the subjects was 10–72 years old. In terms of place of residence, 357 (72%) of participants were from urban areas and the rest were from rural regions (28%; 139/496).

Seroprevalence of anti-*T. gondii* antibodies

Four hundred and ninety six persons were tested for the presence of anti-*T. gondii* IgG and IgM antibodies using ELISA. The overall prevalence of toxoplasmosis was 43.95% (218/496) in Abadan city. Of these, 188 (37.9%) and 30 (6.05%) samples were identified positive for IgG and IgM, respectively.

Risk factors

During this investigation, we recorded eight risk factors related to *T. gondii* infection. In terms of gender, 55% (139/253) and 7% (18/253) of female were seropositive for IgG and IgM antibodies, respectively. While, these antibodies were identified in 20% (49/243) and 5% (12/243) of male, respectively. The overall prevalence between two genders was statistically significant (62% vs. 25%). In terms of residence, 39.2% (140/357) and 56.1% (78/139) of

the subjects of urban and rural communities were seropositive for *T. gondii*, respectively. We divided all participants into four groups including 10–25, 25–40, 40–55, and 55–72 years and the exposure to *T. gondii* was estimated as 44.9% (58/129), 58.05% (75/129), 35.43% (44/124), and 35.96% (41/114), respectively. Also, the higher seroprevalence rate was observed during spring season (64.5%; 80/124), in subjects with the education level of diploma or lower (51.8%; 176/340), from the persons who had contact with cat (49.3%; 69/140), in individuals who consumed raw/undercooked meat (49.3%; 37/75), and amongst who drink unpurified water (52.45%; 32/61). More details are listed in Table 1.

Discussion

The prevalence rate of toxoplasmosis in the Iranian general population was defined 39% (95% CI = 33–46), ranged from 12% to 87.5% with more endemicity in the northern provinces of the country (Daryani et al. 2014). Our knowledge is poor about the epidemiology of *T. gondii* infection and public health significance of urban and rural communities in Abadan city, southwest of Iran. Hence, the present study is the first survey to assess the seroprevalence rate of toxoplasmosis and related risk factors among human general population who were referred to the Taleghani hospital of Abadan city. Anti-*Toxoplasma* IgG and IgM antibodies were found in 37.9% and 6.05% of participants, and the overall prevalence was determined as 43.95%. The

Table 1 Demographic characteristics and risk factors related to seroprevalence of *T. gondii*

Characteristic	Number tested	IgG positive		IgM positive		Total	
		Number	Percent (%)	Number	Percent (%)	Number	Percent (%)
Gender							
Male	243 (49%)	49	20	12	5	61	25
Female	253 (51%)	139	55	18	7	157	62
Age							
10–25	129 (26%)	50	38.7	8	6.2	58	44.9
25–40	129 (26%)	65	50.3	10	7.75	75	58.05
40–55	124 (25%)	38	30.6	6	4.83	44	35.43
55–72	114 (23%)	35	30.7	6	5.26	41	35.96
Residence							
Urban	357 (72%)	122	34.2	18	5	140	39.2
Rural	139 (28%)	66	47.5	12	8.6	78	56.1
Season							
Spring	124 (25%)	70	37.2	10	8	80	64.5
Summer	124 (25%)	25	13.3	4	3.2	29	23.4
Autumn	124 (25%)	65	34.6	11	8.8	76	61.3
Winter	124 (25%)	28	14.9	5	4	33	26.6
Education level							
Diploma or lower	340 (68.5%)	152	44.7	24	7.06	176	51.8
University degree	156 (31.5%)	36	23.07	6	3.8	42	26.9
Contact with cat							
Yes	140 (28.2%)	60	42.8	9	6.4	69	49.3
No	356 (71.8%)	128	35.9	21	5.9	149	41.8
Consumption of raw/undercooked meat							
Yes	75 (15.1%)	30	40	7	9.3	37	49.3
No	421 (84.9%)	158	37.5	23	5.5	181	43
Source of drinking water							
Purified water	435 (87.7%)	163	37.5	23	5.3	186	42.75
Unpurified water	61 (12.3%)	25	41	7	11.5	32	52.45

seroprevalence of our study was a little higher than the average seroprevalence of the Iranian general population (Daryani et al. 2014). Our findings are in agreement with other studies such as Mostafavi et al. in Isfahan province 41.4% (248/599) (Mostafavi et al. 2011) and Keshavarz et al. in Karzj city 45.5% (Keshavarz et al. 1998). In some studies such as Salahi-Moghaddam and Hafizi in Tehran province (Salahi-Moghaddam and Hafizi 2009) and Shahmoradi et al. in Gilan province (Shahmoradi et al. 1998) seropositivity in human general population were reported more than 50%, while the lower seroprevalence of *T. gondii* infection was reported from Chaharmahal & Bakhtiari province (34.2%) (Manouchehri Naeini et al. 2012), Kermanshah province (36.3%) (Mansouri et al. 2003), Meshkin-Shahr city (18.3%) (Soltan Mohammad Zadeh et al. 2003), Shiraz city (27.6%) (Razavi et al. 2003), and Tabriz city (29.87%) (Fatollahzadeh et al.

2016). The discordance between studies could be justified by study region, study population, environment status, type of sampling, number of participants, cultural habits of the subjects, the methodologies, different cutoff values or antibody titers, etc.

Several behavioral and ecological risk factors are involved in the epidemiological distribution of toxoplasmosis. Many probable risk factors have been determined for acquiring the *T. gondii* infection, however inconsistent results are abundant (Daryani et al. 2014; Foroutan-Rad et al. 2016a, b; Gebremedhin and Tadesse 2015; Mansouri et al. 2017; Rostami et al. 2016; Wang et al. 2018). In this study, eight potential risk factors related to toxoplasmosis were assessed. The ELISA results showed 62% (157/253) and 25% (61/243) of female and male were seropositive for anti-*T. gondii* antibodies, respectively. Based on recent systematic review paper among Iranian general population,

a non-significant association was observed between toxoplasmosis and gender ($P > 0.05$). The authors reported a slightly higher prevalence in male 44.08% (95% CI = 34.5–53.9) than female 42.06% (95% CI = 32.04–52) (Daryani et al. 2014). Besides, the same association was observed from Iranian and Chinese apparently healthy blood donors (Mansouri et al. 2017; Wang et al. 2018).

In order to assess the seroprevalence of toxoplasmosis among different age groups, the participants were categorized into four groups, including 10–25, 25–40, 40–55, and 55–72 years. The seroprevalence rate was 44.9, 58.05, 35.43, and 35.96% in these groups, respectively. In this case, Daryani et al. among the Iranian general population of Iran (Daryani et al. 2014) and Gebremedhin and Tadesse in humans from Ethiopia (Gebremedhin and Tadesse 2015) reported a significant association between *T. gondii* infection and age, while Wang et al. among Chinese blood donors failed to show the same association (Wang et al. 2018). As it was predictable, seroprevalence tended to increase with age. The older subjects have a higher seroprevalence of toxoplasmosis than younger groups. Its reason could be explained due to increased exposure to *T. gondii* via one of the following transmission routes: ingestion of raw/undercooked meat, consumption of raw/unwashed vegetables, contact with contaminated soil, contact with cats, etc. (Daryani et al. 2014; Gebremedhin and Tadesse 2015).

In the current study, *T. gondii* infection was higher in individuals who had a history of contact with cats. Cats are serving as definitive hosts for parasites and due to their close contact with humans, especially in rural regions, there is a general consensus that contact with cat could be a potential risk factor for acquiring the infection. Stray and domestic cats are one of the main sources of *T. gondii* infection. They can release millions of oocysts through feces of infected cats into the environment. Sporulated oocysts may survive for a long time in moist soils, ranged from months to years (Dubey 2008; Foroutan et al. 2018a). Accordingly, exposure to soil could play a main role in *T. gondii* infection. In this case, Cong et al. revealed that exposure to soil is associated with prevalence of toxoplasmosis in the study population (Cong et al. 2015), while Yousefi et al. failed to show the association (Yousefi et al. 2017). As it was said in study area section, Abadan city had proper humidity and average temperature, which are necessary for oocysts sporulation. In Iran, 34% (95% CI = 22–46) of cats were found infected with *T. gondii* during 1975–2013 (Foroutan et al. 2018a). Previously published articles, among the Iranian general population ($P < 0.05$), and Iranian pregnant women ($P = 0.002$) have reported that contact with cat is significantly associated with seroprevalence of *T. gondii* infection (Daryani et al. 2014; Foroutan-Rad et al. 2016a).

Consumption of raw/undercooked meat is considered as a potential risk factor regarding the infection (Belluco et al. 2016). In the present study, the more prevalence rate was seen among subject who consumed raw/undercooked meat (49.3% vs. 43%). Although, in many papers no association between toxoplasmosis and this risk factor was shown (Foroutan-Rad et al. 2016a; Gebremedhin and Tadesse 2015; Yousefi et al. 2017), but some others are believed eating raw/undercooked meat can significantly increase the rate of infection in societies (Daryani et al. 2014; Foroutan-Rad et al. 2016b). Transmission of the parasite through meat consumption depended upon the cultural habits of people in a certain area (Belluco et al. 2016; Daryani et al. 2014). Recently, was reported that 2.6%, 12.3%, and 14.7% of cattle, pigs, and sheep were positive for *T. gondii* using different diagnostic methods from a global perspective. The authors confirmed the role of meat and other meat products as *T. gondii* sources. Increase in consumer knowledge surely influences in reduction of the infection (Belluco et al. 2016).

In the current study, 39.2% and 56.1% of participants from urban and rural regions were seropositive for toxoplasmosis, respectively. The villagers habits such as direct exposure to soil, livestock, pets, etc. could make this condition predictable, as earlier reported in the majority of published articles (Foroutan-Rad et al. 2016a, b; Gebremedhin and Tadesse 2015). In the Iranian general population, risk factor analysis highlighted that by increasing the level of education, *T. gondii* infection can be reduced ($P < 0.0001$) (Daryani et al. 2014). In this study, the same association was observed. As it is evident, lack of awareness about the main transmission routes and sources of infection as well as poor hygienic status are extremely important factors to acquiring toxoplasmosis.

In conclusion, our study revealed a moderate seroprevalence of *T. gondii* in the general population of southwest of Iran. A single risk factor may have a little importance on the seroepidemiological pattern of toxoplasmosis in a certain region on its own, but the above-discussed risk factors can change the distribution figure of infection in different geo-locations. Increase of knowledge of people about toxoplasmosis, the main transmission routes, and the related risk factors surely affects in reduction of the infection, especially in rural communities.

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Authors' contributions S. Soltani, M. Foroutan, H. Afshari, M. Hezarian, and M. Sagha Kahvaz conceived, designed and drafted the manuscript; S. Soltani, H. Afshari, and M. Hezarian involved in data acquisition; S. Soltani, M. Foroutan, and M. Sagha Kahvaz involved in statistical analysis and critically revised the text. All authors read and approved the final version of the manuscript.

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Compliance with ethical standards

Conflict of interest No potential conflict of interest relevant to this article was reported.

Ethical statement This study received the approval from the Abadan School of Medical Sciences Ethical Committee (1396.239). A written informed consent was obtained from adult persons and parent or guardian of children less than 15 years old. Ethical issues (Including plagiarism, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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