Original article

Seroepidemiology and risk factors of toxoplasmosis among children age ranged from 1 to 14 years referred to medical diagnostic laboratories in Southeast Iran

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ABSTRACT

Background: Toxoplasmosis is one of the most common protozoan diseases in both developing and developed countries. Human infection occurred by incidental ingestion of oocyst-contaminated food, water, soil or tissue of raw meat. Studies of infection in Iranian children are infrequent. In this study, we investigated seroprevalence, and risk factors of toxoplasmosis among children under 15 years old in Sistan and Baluchistan province, Southeastern Iran.

Materials and methods: 869 sera samples were investigated for the detection of *Toxoplasma gondii* IgG antibodies by ELISA. Moreover, the participant's data were collected to study their association with infection. Data analysis for this study was performed using the SPSS software version 20.

Results: *T. gondii* IgG antibodies were detected in 16.2% (141/869) of children in this study. Regarding the sociodemographic variables, age, male gender, contact with animal pets and white blood cells (WBC) count were significantly associated with toxoplasmosis ($P < 0.001$).

Conclusion: Our study showed relatively high seroprevalence of toxoplasmosis among children in Sistan and Baluchistan province. Our data considered a vital tool for healthcare authorities. Further investigations are needed in different part of Iran especially among high-risk groups of toxoplasmosis.

1. Introduction

Toxoplasmosis is a major zoonosis caused by the protozoan parasite *Toxoplasma gondii* (*T. gondii*), moreover, it is a global health issue, particularly in warmer developing countries.^{1,2} *T. gondii* belongs to the phylum Apicomplexa which able to infect all warm-blooded animals and has a two-host in its life cycle.^{3,4} In general, ingestion of tissue cysts in infected, eating raw meat or ingestion of oocysts from the soil, water or cat litter and congenital infection occurs unintentionally in humans by the placenta.³ Toxoplasmosis clinical manifestations are related to

the status of the immune system of the host. Most toxoplasma infections are asymptomatic or oligosymptomatic in immunocompetent people.⁵ *T. gondii* infection in immunocompromised individuals such as cancer patients, organ transplant recipients and HIV/AIDS patients can cause repeated attacks of the brain and manifest as encephalitis and may also occur in multiple organs.⁶ In addition, *T. gondii* infections in pregnant women may transmit to the fetus and cause congenital toxoplasmosis and may include hydrocephalus, microcephalus, intracranial calcifications, retinochoroiditis, strabism, blindness, epilepsy, psychomotor and mental retardation, petechiae due to thrombocytopenia, and anemia.^{7,8}

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Fig. 1. Geographic map of Sistan and Baluchistan province in Iran.

Microscopic examination is the first method for the diagnosis by detection of oocysts in human feces but this method isn't beneficial because humans aren't the definite host. Serology is the gold standard for diagnosis of *Toxoplasma*-specific antibodies (IgG or IgM).^{9,10} This study aimed to evaluate the *Toxoplasma gondii* seroprevalence among children under 15 years old in five cities (Zahedan, Iranshahr, Saravan, Khash and Nikshahr) of Sistan and Baluchistan province in Iran. Other associated determinants such as age, sex, location of residence, geophagia and hematological characteristics were also evaluated.

2. Materials and methods

2.1. Study area and population

This cross-sectional study between September 2016 and June 2017 at the Toxoplasma laboratory in Faculty of public health, Tehran University of Medical Sciences was conducted. Our study subjects were children referred to the laboratories of the medical centers of five cities (Zahedan, Iranshahr, Saravan, Khash and Nikshahr) in Sistan and Baluchistan province (Fig. 1). The province is the second largest province in Iran. It is located between 29°49'24"N and 60°86'69"E and is about 1352 m above sea level with desert climate (average annual temperature and rainfall is 22.8 °C and 98.8 mm respectively)[11].

2.2. Sampling strategy and immunoserology

A total of 869 children were enrolled in this study. A structured questionnaire was used to record the potential risk factors associated with toxoplasmosis such as sex, age, contact with dog and cat, location of residence and geophagia or pica. About 2–4 mL of whole blood samples were taken from each participant via venipuncture. The samples were allowed to clot and centrifuged at 1000 g for 3 min in order to the separation of sera.¹¹ The collected sera were transported in ice to the Parasitology Laboratory of Tehran University of Medical sciences, where the sera were stored –20 °C until examination. Sera samples were screened for anti-*Toxoplasma* IgG antibodies using ELISA kit (EUROIMMUN, Germany) based on the manufacturer's instructions. The kit has sensitivity and specificity of > 98%.

2.3. Hematological assessment

Hematological parameters such as total hemoglobin, total RBC, WBC count, and Platelet were examined for each sample. Total RBC and

WBC count was obtained by direct microscopic observation in a Neubauer chamber.

2.4. Statistical analysis

Data analysis for this study was performed using the SPSS software version 20 (SPSS, Chicago, IL, USA). Frequency was used for the description of characteristics of children, prevalence of the parasite, and risk factors. Associations between seropositivity for toxoplasmosis and the potential risk factor were evaluated by Pearson's chi-square test. Probability (P) value ≤ 0.05 was considered as statistically significant in all the analyses.

3. Results

3.1. Patient's characteristics

In period between September 2016 and June 2017, out of 869 children 404 (46.5%) females and 465 (53.5%) were males. The mean age was 6.9 ± standard deviation (SD) with the ranged from 1 to 14 years old. Study subjects were divided into groups with predominance of children in age group 6–10 years (46%). Most of the participants were from the Zahedan city (50.9%, 442/869), and the rest from the Iranshahr city (20.1%, 175/869), Saravan city (10%, 87/869), Khash city (9.8%, 85/869) and Nikshahr city (9.2%, 80/869) [Table 1]. Table 2 summarizes the risk factors for toxoplasmosis among the studied children. Out of total participants, 719 (82.7%) living in rural regions and the remaining (17.3%) living in urban areas. Among the participants 236 (27.1%) had history of contact with domestic animals (dog and cat), whereas pica or geophagia was reported among 75 children (8.7%). Hematological parameters of the participants are summarized in Table 3.

3.2. Seroprevalence and risk factors of *Toxoplasma gondii*

Sera from 16.2% (141/869) of children were positive for anti-bodies against *T. gondii* by ELISA.

Seropositivity was more prevalent among males 99 (70.2%) than in females 42 (29.8%) with significant difference according to gender ($p < 0.05$). The association of *T. gondii* seropositivity and socio-demographic data was statically significant with age and contact with pets (dogs and cats) ($p < 0.05$). Hematological characteristics of the children showed that *T. gondii* seropositivity was associated with white

Table 1

Seroprevalence of *Toxoplasma* infection among children referred to the laboratories of medical centers in five cities of Sistan and Baluchistan province, Southeastern Iran (n = 869).

	Samples N(%)	Positivity N(%)	Prevalence Ratio	P value*
Variable (City of residence)				0.183
Zahedan	442 (50.9%)	69 (48.9%)	1	
Iranshahr	175 (20.1%)	23 (16.3%)	0.8	
Khash	85 (9.8%)	14 (9.9%)	0.9	
Nikshahr	80 (9.2%)	16 (11.4%)	1.3	
Saravan	87 (10%)	19 (13.5%)	1.4	
Total	869 (100%)	141 (100%)		

blood cells (WBC) ($p < 0.05$). The seroprevalence in Zahedan city was slightly higher than that of other cities, although not statistically significant ($p = 0.183$). ($p > 0.05$).

4. Discussion

Toxoplasmosis is one of the most common protozoan zoonotic diseases worldwide. *T.gondii* infection is mostly asymptomatic for months to years.^{12,13} The main potential factors associated with *T. gondii* infection are socioeconomic, geographical and demographical and cultural differences, such as living and eating habits of the population.^{14,15} Based on the Human Development Index (HDI), Sistan and Baluchistan province has poor ranking among other provinces in Iran especially in terms of life span, education level and income.^{16,17} Consequently, the province is plunged with parasitic infections; toxoplasmosis inclusive. In the present study, an overall anti-toxoplasma antibodies seroprevalence of 16.2% among children of Sistan and Baluchistan province were reported. The prevalence toxoplasmosis in children under 15 years old had a significant correlation with the age, sex, contact animal pets and hematological parameters such as the number of WBC. A meta-analysis reported that the seroprevalence of *Toxoplasma gondii* infection among the Iranian general population is 39.3%,¹⁸ the higher prevalence rate was reported in Iranian cities such as Gilan (86.3%)¹⁹ and Isfahan (41.4%).²⁰ Our finding showed similar prevalence which reported in Shiraz (18.26%)²¹, while lower prevalence was reported in Lorestan (4.4%).²² Other studies in Tehran and Mazandaran reported higher prevalence than our study, but lower than the national seroprevalence.^{23,24} The Seroprevalence rate of *Toxoplasma gondii* infection was showed a wide range from 4 to 84% depending on the environmental and socio-economic conditions from different parts of the world.²⁵ However, the seroprevalence of toxoplasmosis in the present study is lower than reported in Mexico (49%),²⁶ Libya (45%),²⁷ and

Netherlands (35%).²⁸ On the other hand, our seroprevalence rate is higher than that reported in South Korea (9.5%),²⁹ in agreement with studies in Singapore (17%)³⁰ and Sweden (18%).³¹ The variation in the prevalence rate of infection could be related to variation in weather conditions, nutritional status, location and culture of the population.³² In this study, the prevalence of *Toxoplasma* infection among males (70.2%) was significantly higher than among females (29.8%) probably because of the gender-specific characteristics, such as outdoor activities and sports among boys which increase the risk for *Toxoplasma* infection. Our results were similar to those reported from Tehran³³ and Isfahan³⁴ which showed higher prevalence among males. The current study showed that the age group 6–10 years is a higher risk than other age groups without understanding the reason of that to us. Domestic animals (especially the cats), as the definite host of toxoplasma, play an important role in the transmission of infection to humans. Generally, the transmission of the infection is accentuated by contact with domestic animals (pets). Our study showed that contact with pets is the main risk factor for *Toxoplasma* infection. Our study findings were similar to those reported in Lorestan.²⁴ In this study, a significant association was observed between WBC counts and *Toxoplasma* infection, WBC count was increased against *T.gondii* infection.³⁵ In our study, no significant association was found between pica and *Toxoplasma* infection. However, pica or geophagia increases the risk of toxoplasmosis, especially among children living in homes with pets or children playing with soil, as reported in a number of studies.^{36–38} A high prevalence of *Toxoplasma* infection has been reported among nomads, probably due to their low quality of life and adversities of climatic conditions.³⁹ Our study also showed that children from rural areas were more susceptible to infection than those who live in urban areas, but this difference wasn't statistically significant. A similar finding was also reported in Isfahan.³⁴ In southwestern Iran, the prevalence of toxoplasmosis had been reported about 24% in individuals with hydatidosis, which was

Table 2

Seroprevalence of *Toxoplasma* infection among children referred to the laboratories of medical centers in Sistan and Baluchistan province, according to Demographic characteristics (n = 869).

	Samples N(%)	Positivity N(%)	Prevalence Ratio	P value*
Variable(Age in years)				0.001*
1-5	347 (40%)	23 (16.4%)	1	
6-10	400 (46%)	70 (49.6%)	1.2	
11-14	122 (14%)	48 (34%)	3.2	
Variable(Sex)				0.001*
Male	465 (53.5%)	99 (70.2%)	1	
Female	404 (46.5%)	42 (29.8%)	0.55	
Variable(Contact with dog and cat)				0.001*
Yes	236 (27.1%)	85 (60.3%)	1	
NO	633 (72.9%)	56 (39.7%)	0.25	
Variable(Pica or Geophagia)				0.962
Yes	75 (8.7%)	12 (8.6%)	1	
NO	794 (91.3%)	129 (91.4%)	1.01	
Variable(Location)				0.627
urban area	150 (17.3%)	22 (15.6%)	1	
rural area	719 (82.7%)	119 (84.4%)	1.1	

*p = < 0.05, difference is statistically significant.

Table 3

Seroprevalence of *Toxoplasma* infection among children referred to the laboratories of medical centers in Sistan and Baluchistan province, according to their hematological characteristics (n = 869).

Lab results	Samples N(%)	Positivity N(%)	Prevalence Ratio	P value*
Variable(Hemoglobin)				0.900
Normal	680 (78.2%)	111 (78.7%)	1	
Low	189 (21.8%)	30 (21.3%)	0.98	
Variable(WBC count)				0.001*
Normal	764 (87.9%)	108 (76.6%)	1	
high	105 (12.1%)	33 (23.4%)	2.2	
Variable(RBC count)				0.596
Normal	806 (92.7%)	129(91.5%)	1	
Low	63 (7.3%)	12 (9.5%)	1.2	
Variable(platelet)				0.648
Normal	791 (91%)	130 (92.2%)	1	
Low	78 (9%)	11 (7.8%)	0.85	

*p = < 0.05, difference is statistically significant.

higher than the study population.⁴⁰

5. Conclusion

Our results documented that the seroprevalence of *Toxoplasma* infection among children of Sistan and Baluchistan province revealed that 16.2% infected and the infection is statically significant associated with male gender and contact with pets. This information can be a useful tool to the health authorities in order to offer proper health education programs with a view to the prevention of toxoplasmosis especially to the sub-population at risk. Further investigation involving other high-risk groups of toxoplasmosis such as pregnant women, transplant recipients, cancer and HIV/AIDS patients, in this and other provinces of Iran is suggested.

Author's contribution study concept and design

VR, SE, FB,AT,acquisition of data: VR, OR,AI, MG,ZH,SE analysis and interpretation: VR, GA, RA,MKS.

Declaration of competing interestCOI

The authors and coauthors declare that they have no conflict of interest that affects this study.

Ethical statement

The Ethics Committee of the Iranshahr University of Medical sciences (Permit Number: IR. IRSHUMS.1394.10), approved this research. Parents of all the children included in the study were verbally informed about the study's aims, and collection of the specimens was done after their consent was obtained.

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