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SEROPREVALENCE OF IGG AND IGM TO TOXOPLASMOSIS AMONG PREGNANT WOMEN IN SOUTH-EAST OF IRAN

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Abstract

Toxoplasmosis is one of the most prevalent zoonotic diseases worldwide that following the consumption of raw or half-cooked or through contamination with infected cat feces, as well as congenital Toxoplasma gondi is transmitted through the placenta to the fetus. In pregnant women, Toxoplasmosis is mostly without any symptoms and in warm climatic conditions usually more common than cool climatic and mountainous. The serological techniques are most important methods for diagnosis. This study was a cross - sectional that 185 sera samples were collected from pregnant women referring to reference laboratory clinic of Nikshahr district in 2012 and the IgG and IgM Antibody levels against Toxoplasma in their sera were examined using ELISA method.10.3% cases out of all samples were IgG positive and in 0.55% cases IgG and IgM both positive for Toxoplasmosis. Among participants in this survey 98.9% had the history of consuming raw vegetables, 9.7% with the history of consuming half-cooked meat, 78.9% with the history of not using disinfectant materials in washing vegetables, 5.4% had the history of contacting cats and 16.8% had consumed contaminated water. This study for the first time demonstrated 89.7% of pregnant women in Nikshahr district were serologically negative against IgG Toxoplasma antibody. However, such community that given the low level of safety and risks of infection during pregnancy and its complications, the public education system, health care is important to prevent infection.

Keywords: Toxoplasmosis, IgM, IgG , Pregnant women, South-East, Iran.

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INTRODUCTION

Toxoplasma gondii is an obligate intracellular protozoan parasite that has a global distribution in animals and human. Cat as the main host and other mammals and birds as intermediate hosts serve as source of infection of this protozoan (Lee *et al.,* 2008; Dubey, 1998). This agent is transmitted mainly through the ingestion of oocysts excreted in the feces of infected cats or by meat from an intermediate host containing cysts. Cats can be used as sentinels for the environmental spread of T. gondii in densely built urban areas because they are exposed to all the infective forms of the parasite without any protection (Meireles *et al.,* 2004). Infection with this protozoan usually occurs by the ingestion of raw or undercooked infected meat, vegetables and water contaminated with its oocyst, and through the placenta (Trans placental transmission) from an infected mother to her fetus, and via tachyzoites contained in blood products, tissue transplants, and unpasteurized milk (Pelloux *et al.,* 1997). It is particularly dangerous for pregnant woman transmitted the disease to their fetuses and newborns (Borna *et al.,* 2013), and also for the immunocompromised hosts such as AIDS patients (Dubey *et al.,* 1998). In severe cases, they can be affected by encephalitis, myocarditis, and chorioretintis or finally die. Most pregnant women with acute acquired infection do not experience obvious symptoms or signs. A minority may experience malaise, low-grade fever, and lymphadenopathy (Lin *et al.,* 2008). Severe clinical signs in the infected infant are more commonly observed in offspring of women whose infection was acquired early in gestation. In case of infection in first trimester incidence of trans placental infection is low15%, but disease in neonate is most sever, but in case of infection in third trimester incidence of trans placental infection is high 65% but infant is usually asymptomatic at birth (Montoya and Liesenfeld, 2006; Skallova *et al.,* 2006). The prevalence of T. gondii differs among countries worldwide, also within a country. It has been estimated that up to one third of the world's population has been infected with endemicity from around 10% to 70% (Alvarado-Esquivel *et al.,* 2009). Totally Serological positive cases increase by rising in age and usually after the age of 25 in both genders are the same (Edrisian, 1990). Obvious differences among prevalence of toxoplasmosis in different areas are because of geographical

climate dispersal or diet and alimentary habits, are on alimentary factors, it is because in dry areas toxoplasmosis is less prevalence than in hot and damp areas (Boyer *et al.,* 2005). Prevention of congenital toxoplasmosis in pregnant women has been based on serological Test for Toxoplasma antibodies. Many serological tests including the Latex Agglutination test, ELISA, indirect fluorescence antibody tests (IFA) and Haemagglutination test have been utilized in the detection of antibodies against T. gondii (Song *et al.,* 2005).

There have been several reports regarding the screening of *T.gondii*antibodies among Iranians. The rate of toxoplasmosis in Iranian cities was reported in 2008 By Iranian Ministry of Health. The results were accordingly 40.7% for Isfahan, 44.2% for Lorestan, and 34.2% for Bandar-e-Abbas (Daryani, 2004; Mohammadi *et al.,* 2008). In a serologic study in Zahedan among 337 individual samples of pregnant women using indirect IFAT method the prevalence of specific antibodies were 49.8% in 1378 (Mahdi, 2001). In another study in Chaharmahal and Bakhtyari the seroprevalence of Toxoplasma antibodies among pregnant women using IFA was 27.6 % (Ebrahimzadeh *et al.,* 2013). As 90% of patients or more have no any sign and it is possible that doctor could not distinguish toxoplasmosis in these women. Denoting and measuring prevalence of anti-toxoplasma anti bodies is a useful step in different parts of Iran before pregnancy.Nikshahrdistrat is a tropical region in southeast of Iran. There is scarce information about the epidemiology of *T. gondii*infection in pregnant women in Nikshahrdistrict. This study performed in order to determine the *Toxoplasma* antibodies in pregnant women in Nikshahrdistrict, South-East of Iran using ELISA method.

MATERIALS AND METHODS

In this cross sectional study the sample size was calculated as 185 cases in 2012.The samples were pregnant women referring to the reference laboratory of Nikshahr district for routine pregnancy tests. Inclusion criteria for the study subjects were pregnant women, of all ages and at any stage of pregnancy, residing in Nikshahr district. Epidemiological data, including socio-demographic and behavioural characteristics, were obtained from all pregnant women. Socio-demographic Characteristics included age, place of residency, and educational and socio-economic levels. Behavioural characteristics included cat contacts, consumption of raw or undercooked meat, contaminated water, and unwashed raw vegetable or fruit consumption. Clinical characteristics encompassed of pregnancies and abortions. Clinical data were obtained during a direct interview with the participants.

A total of 185 serum samples were tested at the Department of Parasitology, School of Medicine, Zahedan University of Medical Sciences, Iran. Blood samples were collected and sera separated by blood centrifugation at 3000 rpm for 5 min and frozen at -20 °C until use. Sera of all pregnant women were assayed for T. gondii antibodies using commercially available enzyme immunoassay kits. IgG antibodies were assayed using a ‘‘Pishtaz Teb Toxoplasma IgG, Iran’’ kit. Sera positive for T. Gondii IgG antibodies were further tested for T. Gondii IgM antibodies with a ‘‘PishtazTeb Toxoplasma IgM, Iran’’ kit. Both Tests were accomplished according to the manufacturer’s manual procedures. The statistical analysis was performed using SPSS version 18.0.

We used the chi-square and the Fisher’s exact test for comparison of frequencies among groups.

RESULTS

Out of 185 pregnant women included in this study.35 individuals (18.9%) were living in town and others in villages, they were at least 17 years old at most 37, most abundance were in group of 17-23 years (44.9%) and least abundance in group of 31-37 years (13.5) and 45 individuals (24.3%) were illiterate, 121 (65.4%) diploma and 19 (10.3%) were above diploma. Out of all surveyed cases 89.7% were IgG negative and 10.3% were IgG positive and 0.55% were IgM positive, and 99.45% were negative for IgM, Out of all of them just 0.55% of samples were positive for IgG and IgM simultaneously (Table 1).

The Survey results showed that abundance of positive results seen in group of 31-37 years (21.5%).By using statistical tests we didn’t observe significant difference between age group of positive IgG and negative IgG. None of them experienced abortion or exposure to cat. Positive IgG individuals used half-cooked meat but in negative IgG users were 57.5% .comparison survey using statistical test between two groups showed no significant relations. 5.26% of positive IgG individual used half cooked meat that in people with negative IgG it was 10.3Comparative survey using statistical test between two groups did not show signification relation.63.1% of positive IgG didn’t use washing materials or disinfectants to wash vegetables whereas in people with negative IgG it was 83.13% Significant difference ($p<0.05$) was observed in of cases. (Table.2). Results showed that abundance of dangerous behaviour in people with positive IgG was 5.26% using raw meat,61.3%,not using disinfectants for washing vegetables and 22.2% using pit water consecutively (Figure1).

DISCCUSION

Toxoplasmosis is a zoonotic infection with worldwide distribution and exhibits the wide range of prevalence based on sociocultural and climatic conditions (Fazaeili and Ebrahimzadeh, 2007). Toxoplasmosis is an important disease in pregnant women and organ transplant and immunodeficient patients (Barbosa *et al.,* 2009). Congenital Toxoplasmosis is a major problem in most communities with a high prevalence of T. gondii infection and study of the seroepidemiology of this infection among women of childbearing age could provide appropriate approaches to design the preventive measures (Daryani, 2004). The present study showed a sero prevalence of 10.3% among 185 pregnant women. Therefore these pregnant women were not at risk for toxoplasmosis. The results show a dramatically decrease in the prevalence of sero positive individuals in comparison to Ebrahimzadeh *et al.,* results using Elisa and PCR method in 2013 among pregnant women in Zahedan (17,18). The epidemiology of T. gondii infection in pregnant women had not been studied yet in Nikshahr district. Most studies in Iran have been focused on childbearing age and pregnant women and wide range of prevalence rate of Toxoplasma antibodies have been reported as follows: Sari 74.6%, Tehran 31%, Kerman 29.4% (Daryani, 2004; Iqbal *et al.,* ?) ,Sanandaj 28.2% and Hamedan 33.5% (Mohammadi *et al.,* 2008).

Table 1.The correlation between IgG and IgM levels among studied individual

		IgM Negative	IgM Positive
IgG Negative Individuals	Cases	166	0
	Percent	89.7	0
IgG Positive Individuals	Cases	19	1
	Percent	94.73	5.26
All Studied	Cases	185	1
	Percent	99.45	0.55

Table 2.The Frequency Distribution of Specific anti-Toxoplasma IgG Positive among Pregnant Women According to Examined Variables

	IgG Negative Individuals Percent(Cases)		IgG Positive Individuals Percent(Cases)	
	Percent	case	percent	case
Cat Contact History	yes	10	0	0
	no	156	100	19
Raw Vegetable Consumption	yes	166	10.3	19
	no	0	0	0
Half-cooked Meat Consumption	half-cooked	149	94.7	18
	cooked	17	5.3	1
Age Groups	17-23	77	31.6	6
	24-30	68	47.4	9
	31-37	21	21.1	4
Washing Vegetables using disinfectants	no	138	63.2	12
	yes	28	36.8	7
drinking water habits	Piped Water	137	89.5	17
	well Water	29	10.5	2
Education Level	No educated	40	26.3	5
	elementarily	42	31.6	6
	High school	34	15.8	3
	diploma	35	5.3	1
	Diploma up	15	21.1	4
Place living	urbane	27	42.1	8
		139	57.9	11

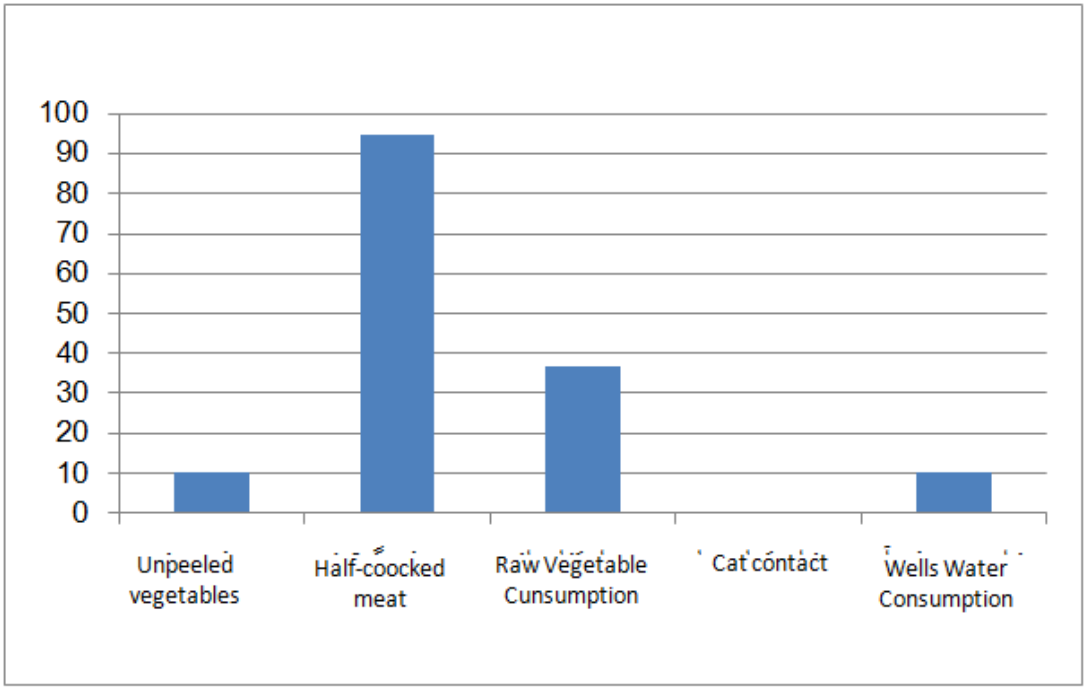

Figure 1. The rate of Risk Factors among IgG Positive Individuals

Several studies in different countries and cities around the world have indicated that seroprevalence rate of Toxoplasma antibodies have a wide range as follow: Brazil 66.3% (18) India 49.52% (Fallah *et al.,* 2005), Thailand 28.3% (Sarkar *et al.,* 2012), Turkey 49.4 (Nissapatorn *et al.,* 2011), and France 43.8% (Al-Mendalawi, 2010). This study was performed on pregnant women to distinguish previously infected women from women who had not been previously infected and to identify the prevalence rate of pregnant women at risk of Toxoplasmosis. In this study, 19 out of 185 cases (10.3%) were positive for specific IgG antibody and 1 cases (0.55%) were positive for specific IgM antibody, therefore, this study showed a moderate seroprevalence (10.3%) of T. gondii IgG antibody compared to other parts of the Islamic Republic of Iran (Daryani, 2004; Mohammadi *et al.,* 2008; Iqbal and Khalid, ?).

Results of this study indicate that the prevalence of Toxoplasma antibodies in Nikshahr district is lower than some other parts of Iran. Although, Nikshahr district climate is warm and arid but the mean temperature of Nikshahr district during the year is higher than other area such as the central, north and the west parts of Iran and this temperature is not optimum for sporolation of Toxoplasma oocysts. Results from some studies have indicated that the risk of T.gondii infection increased with age (26). Although, there was no association between seropositivity of Toxoplasma antibody and different age groups in our study, but the seropositivity rate in 24-30 age group was obviously higher than other groups. This result is in accordance with the results of some studies (Alvarado-Esquivel *et al.,* 2009; Daryani, 2004; Iqbal and Khalid, ?).

Results of this study confirm the results of above mentioned studies. So promoting level of education is one of the variables that could effect on 'hygienic behaviors. In this study we saw a significant relation between birth place and habitat with positive serology and people in rural areas had more positive amounts than urban areas that are probably because of more exposure to cat and other domestic animals and also using raw vegetables and not washing vegetables properly. In some study in Gorgan there wasn't any difference between town and village women (20). In this study 5.3% of individuals with positive IgG used raw meat, that is similar with study done by Kayabi and Atai that 3.44% of individuals with positive IgG used half cooked meat. Results of this study showed that all people with positive IgG used vegetables also 63.2% of individual with positive IgG didn't use antiseptic for washing. Obviously the main way of transferring contamination in this area is by ingestion of parasite via eating vegetables that is because of wrong ways of washing vegetables. Nikshahr district is a tropical region in southeast of Iran and the drinking water is supplied from 200 kilometer away from Zabol City by pipelines. Almost all households in Nikshahr district have a metallic reservoir for reserving the water, which can increase the chance of contamination of drinking water. Vegetables are also provided from other cities and can be contaminated during transportation. Altogether, results of this study indicates that 65% of pregnant women in Nikshahrdistricti do not have immunity against Toxoplasmosis, therefore they are at risk of congenital Toxoplasmosis for their fetuses and it is necessary for health policy makers to design the preventive measures against congenital Toxoplasmosis.

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