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Original Article

Comparison of Toxoplasmosis Seroprevalence in Multi Partner, Married (Monogamous) and Virgin Iranian Women

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Abstract

Background: Toxoplasmosis could lead to serious outcomes during pregnancy. The aim of this study was to investigate serologic toxoplasmosis in three groups of women according to number of sexual partners.

Methods: The frequency of anti-*Toxoplasma* IgG from 471 women (101 virgin girls, 240 married women and 130 multi partner women) were determined by ELISA method from referred participant to medical centers of Tehran, Iran in 2020. The results were analyzed by chi-square and logistic regression tests.

Results: Prevalence of toxoplasmosis was significant with the number of sexual partner according to chi square test (P<0.001) and the highest one was observed in multi partners' women (56.2%) and the lowest one in virgin girls (17.8%). ORs of virgin girls and multi partners' women were 0.594 and 3.758 respectively, compared to married women. The effect of age on the frequency of anti-*Toxoplasma* IgG in married women was significant but it was not significant in multi partners' women. In addition to IgG frequency in married women and multi partners' women had no significant relationship with the number of children.

Conclusion: Having sexual activity after marriage and having multi partner in sexual activity may possibly be a novel risk factor for toxoplasmosis infection or increasing the IgG frequency.

Introduction

Toxoplasma gondii is the most common parasitic infection between humans and other vertebrates, reported with various outbreaks in almost all countries worldwide

(1). The high levels of antibody titer against *Toxoplasma* were reported between 16% and 40% in the North American and British communities and between 50% and 80% in



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South America, Central America, and the Europe (2). Moreover, these levels vary between 18% and 70% in other countries (3).

The most important way of transmitting human infections is from mother to fetus's transmission; eating raw meat infected with parasitic cysts; and eating water, food, and vegetables contaminated with oocytes (4). Although the infection in healthy people is usually benign and asymptomatic (5), these protozoa can cause encephalitis as well as death in people with immunodeficiency (6).

The transmission of toxoplasmosis through sexual contact was reported in some animals such as sheep and goats. The protozoan tachyzoites were found in the sperm of these animals and the infection was seen in their offspring; the tachyzoite of this parasite was also isolated from human sperms, so sexual contact can be suggested as another possible transmission way (7-11).

We aimed to determine and compare the prevalence rates of toxoplasmosis in women with different numbers of sexual partners.

Materials and Methods

This cross-sectional study conducted with ethics approved ID: IR. S BMU.MSP.REC.1397.481 in Tehran City in 2020.

A total of 471 women were assessed in terms of number of sexual partner and toxoplasmosis infection in four urban health centers with different socio-economic statuses. Most of the women come to these centers for usual health care and maternal care; one of these centers has a triangular clinic designed for special care for multi partners' women (the women who have more than one sex partner). A blood sample (5 ml) was collected from each participant and then centrifuged. The serum samples were stored at -20 °C and examined for IgG antibodies against T. gondii using ELISA kit (PISHTAZTEB DIAGNO-SIS Co limited) with 100% sensitivity and 97% specificity. Thereafter, antibody levels were expressed as International Units (IU)/ ml, and the results $\geq 1/1$ IU/ml were considered as positive.

Ethical Approval

This project was approved by Medical School of SBMU with approved ID: IR.S BMU.MSP.REC.1397.481. All participants provide written consents

Variables

Based on number of sexual partner, women were divided to three groups (virgin girls, married women and multi partner women). According to Morgan's sampling table, we needed at least 100 women in each group of this study. Finally, 130 multi partners' women, 240 married women (married women with only one sexual partner), and 101 virgin girls (virgin girls refer to those women who had not married before and declared that they had no sexual partner so far) were volunteer to participate.

The next variable was age, because of the importance of which and its effect on the frequency of anti- *Toxoplasma* IgG, we considered the women in two parts as follows: first, women who were divided in 10 groups with 4 years apart from less than<18 and more than >50 years old. Second, every three women group was divided in 2 parts less than 30 and over than 30 years old, to consider the effect of age on increasing the frequency of IgG in each group separately.

The other variable was having children. In this regard, we divided married women to 5 groups who had no child to 4 children or more, to consider that whether the number of children could increase IgG frequency or not. Having pet (yes or no), washing vegetables with detergent (yes or no), and consumption of semi-cooked meat (yes or no) were the considered items.

Data analysis

The collected data were analyzed using Chisquare, and logistic regression test.

Results

The prevalence rate of toxoplasmosis was significantly associated with the number of sexual partners according to chi square test (P < 0.001). Accordingly, the highest rate was observed in multi partners' women (56.2%) and the lowest one in virgin girls (17.8%) (Table 1).

 Table 1: Frequency of anti-Toxoplasma IgG according to number of sexual partners in 3 groups of women referring to health centers of Shahid Beheshti University of Medical Sciences, Tehran, Iran.

	Groups	IgG1-	IgG+	Total
Virgin	No	83	18	101
	0⁄0	82.2	17.8	100.0
Married	No	178	62	240
	0⁄0	74.2	25.8	100.0
Multi part-	No	57	73	130
ner		43.8	56.2	100.0
Total	No	318	153	471
	%	67.5	32.5	100

We considered the effect of age on the IgG frequency in 3 groups of women aged between 18 and 50 years old and it was also revealed that increasing the frequency of IgG every 4 years with (P<0.001) was significant in all groups. The frequency of anti-*Toxoplasma* in married women and multi partner respectively was 61 and 73 but in virgin girl was just 18 cases, but it needs reconsideration to accept the age effect on frequency of *Toxoplasma* IgG (Table 2).

Table 2: Frequency of anti-*Toxoplasma* IgG in every 3 women groups according age, less than <18 to more than>50 years old, referring to health centers of Shahid Beheshti University of medical sciences

Age group(yr)		Ma	rried	Virgin		Multipartner	
		IgG-	IgG+	IgG-	IgG+	IgG-	IgG+
<18		_	-	2	_	_	-
18-22		20	3	11	1	2	2
22-26		39	4	15	-	3	1
26-30		44	7	16	5	10	9
30-34		41	20	24	5	5	14
34-38		16	14	5	-	9	10
38-42		8	6	4	4	7	6
42-46		4	3	2	-	9	10
46-50		3	2	3	1	9	5
+50		4	2	1	2	3	16
Total	No	179	61	83	18	57	73
	%	74.6	25.4	83.2	16.8	43.8	56.2

We divided each age group into 2 parts, less than 30 and more than 30 years old for better analyses the effect of age on the frequency of anti-*Toxoplasma*. The IgG frequency of all participants under 30 was less than over 30 years old. Although in virgin and multi partners group, it was not significant, but in married group it was significant with (P<0.001) (Table 3).

	Age group (yr)	IgG - I	IgG +	Total
Virgin	Age less than 30	43	7	50
U	%	86.0	14.0	100.0
	Age more than 30	40	11	51
	%	78.4	21.6	100.0
Married	Age less than 30	103	14	117
	%	88.0	12.0	100.0
	Age more than 30	75	48	123
	0/0	61.0	39.0	100.0
Multi	Age less than 30	15	12	27
partner	0/0	55.6	44.4	100.0
	Age more than 30	42	61	103
	0⁄0	40.8	59.2	100.0
Total	Age less than 30	161	33	194
	0⁄0	83.0	17.0	100.0
	Age more than 30	157	120	277
	0/0	56.7	43.3	100.0

Table 3: Frequency of anti-*Toxoplasma* IgG in three women age groups, which divided in 2 parts sub group to consider the age effect on increasing the IgG in every group separately.

Logistic regression test was used to determine the relationship between the abovementioned variables, each of which had increased antibody titer. The results of multivariate analyses cleared that the Frequency of anti-*Toxoplasma* IgG of virgin girls and multi partners' women was correlated with that of married women. The chance of anti-

Toxoplasma IgG increase in virgin girls was 0.594 fold less than married women, but the multi partners' women were 3.758 time more than married women with (significant level of (P < 0.001)). ORs of virgin girls and multi partners' women were 0.594 and 3.758 respectively, compared to married women (Table 4).

 Table 4: logistic regression module for association of having sexual partner with Frequency of anti-Toxoplasma

 IgG in 3 groups of women referring to health centers of Shahid Beheshti University of medical sciences

Variable	В	S.E.	Wald	df	Sig.	Exp(B)
Married			46.568	2	.000	
Virgin girl	521	.304	2.929	1	.087	.594
Multi partner	1.324	.231	32.933	1	.000	3.758
Constant	-1.077	.148	52.724	1	.000	.341

B: beta, SE: standard error, Wald: wald statistics, df: degree of freedom, Sig: significant, Exp: exponential B

Due to the different effect of age and number of children on frequency of anti-*Toxoplasma* IgG, we decided to evaluate them in married women and multi partner women separately. The frequency of anti-*Toxoplasma* IgG in married women with was 4.123 fold, so it is cleared that increasing age effect on increasing frequency of anti-*Toxoplasma* IgG with (significant level of P<0.001). The second variable

was the number of children, which was had a relationship with the anti-*Toxoplasma* IgG, according to multi-variant analyses. With born

of each child, 1.2-fold frequency of anti-*Toxoplasma* IgG increased in married women but it was not significant (Table 5).

 Table 5: Logistic regression module for association of having children, and age groups less than 30 and more than 30 yr with frequency of anti-*Toxoplasma* IgG in married women referring to health centers of Shahid Beheshti University of Medical Sciences, Tehran, Iran

Variable	В	S.E.	Wald	df	Sig.	Exp(B)
Child	.184	.166	1.227	1	.268	1.202
Married women						
	1.417	.354	16.010	1	.000	4.123
Constant	-2.091	.300	48.471	1	.000	.124

B: beta, SE: standard error, Wald: wald statistics, Df: degree of freedom, Sig: significant, Exp: exponential B

We evaluated The frequency of anti-*Toxoplasma* IgG in multi partner women similar the above analyses, which was 1.288 fold, but it was not significant so it was clear that, increasing age had no effect on increasing the frequency of anti-*Toxoplasma* IgG in multi partner women. The second variable was the number of children, which was 1.32 fold more chance to increases the frequency of anti-*Toxoplasma* IgG, but according to multi-variant analyses it was not significant too (Table 6).

 Table 6: Logistic regression module for association of having children and age groups, less than 30 and more than 30 with Frequency of anti-*Toxoplasma* IgG in multi partner women referring to health centers of Shahid Beheshti University of Medical Sciences, Tehran, Iran

Variable	В	S.E.	Wald	Df	Sig.	Exp(B)
Child	.283	.145	3.815	1	.051	1.327
Multi partner women						
	.253	.470	.289	1	.591	1.288
Constant	393	.399	.967	1	.325	.675

B:beta ,SE: standard error , Wald: wald statistics, Df: degree of freedom,Sig: significant, Exp: exponential B

Other variables such as occupation, history of abortion, contact with pets, washing vegetables, and meat consumption were also considered in this study, but no significant relationship was found between the frequency of anti-*Toxoplasma* IgG and these variables (Table 7).

 Table 7: Logistic regression module for some variable, which is not in equation with IgG titer against Toxoplasma gondii in three group's women referring to health centers of Shahid Beheshti University of Medical Sciences

Variables	Score	Df	Sig.
Consumption Semi cooked meat	.217	1	.642
How to wash vegetables	.012	1	.912
Occupation	.235	1	.628
Education level	.017	1	.897
Having pet	1.267	1	.260
Overall Statistics	1.801	5	.876

Score; concession, Df: degree of freedom, Sig: significant

Discussion

The present study was conducted to evaluate the frequency of anti-*Toxoplasma* IgG versus the variables in three groups consisting of virgin girls, married women, and multi partners' women, and it was such as the studies of Montoya et al (12) and Sensini et al (13).

In this study, out of 240 married women, 62 participants (25.8%) had anti-*Toxoplasma* antibody titers, which was less than Dalimi et al (14) research results in pregnant women (26.3%), Hazrat-e-Tappeh et al (15) (28.3%), Naeini et al (16) (37.3%), Hosseini et al (17) (58.8%), Kalantari et al (18) (60.6%) and Abdi.et al (19) (44.8%). However, it was higher than studies conducted by Jones et al (20) and the prevalence rates reported by toxoplasmosis (0.88), Norouz Larki et al (21) (8.9) or Wei Cong et al (22)(15.2) in pregnant women (20-22).

Although the prevalence of *Toxoplasma* in the married women from Tehran City was reported 25.8%, but in the northern cities of Iran with more humidity and high temperature, it was reported as 60.6%. This theory suggests a decreased prevalence (8.9%) in relatively dry and warm provinces such as Tehran (21).

Out of 101 virgin girls, a total of 18 participants (17.8%) who had no sexual activity before, had anti-*Toxoplasma* antibody, which was higher than Sharif Maraghi study (23) reporting an increase in the prevalence of this parasite (12.8%) as well as Modrek study(24) (10%) in virgin girls (24). Notably, it was less than the results of Mahmoodi et al(25) who declared the prevalence of *Toxoplasma* in virgin girls (18.4\%), Amirkhani et al (26) (21.2\%) and Mohammadi. et al (27) (28.2\%) . Accordingly, these differences can be due to health, nutritional, and cultural conditions (25-27).

Comparing this study to other studies indicated a significant difference between the prevalence rates of Toxoplasmosis in virgin girls 0.594 fold less than married women and multi partner women 3.758 time more than married women did. Correspondingly, regarding the fact that other variables are constant, this difference can be attributed to sexual communication. Bitaraf et al (28) conducted a study on the prevalence of Toxoplasma in men and women, which referred to a hospital. Results in different age groups were collected as follows: Women aged under 21 years old (11.01%), between 21 and 30 years old (18.93%), between 31 and 40 years old (28.3%), and older than 40 years old (53.12%), which are similar to the results of this study (28).

In the previous studies, the increased titer was seen in old ages without any justification for them, so, they declared that the common variables was the agent of the increase, but Ilana et al. (29) examined 2109 women aged between 40 and 69 years old in terms of sexual activity. Accordingly, 75% of the women had sexual activity. Moreover, aging in women and the passing of menopause do not lead to loss of libido. Therefore, probably one of the reasons for the increased titer beyond 45 years old is sexual activity (29).

Of the 130-multi partners' women, 73 participant (56.2%) had the frequency of anti-*Toxoplasma* IgG higher than that of virgin girls and married women. Given the fact that other variables were constant for all the groups and only the variable in the number of sexual partner in groups was different, the difference between the frequency and the number of the increased sexual activity in the individuals probably could be considered as the reason.

We used logistic regression for performing more accurate evaluation and determining the OR chance of the contamination occurrence, according to which the multi partner group has more than 3.758 times chance of being infected with this parasite (OR=3.758) compared to married women (P<0.001). However, the virgin girl chance for being infected to *Toxoplasma* is 0.594 fold less than married women. Correspondingly, this is consistent with the single study of *Toxoplasma* on 136 SW and 272 non-SW women, as controls, in Mexico (OR=4.05) (11).

In addition, some other variables such as age and having children as well as their effects on the frequency of anti-*Toxoplasma* IgG have been evaluated in this study.

Sharifi et al (30) examined the seroepidemiology of toxoplasmosis in 1832 men and women referred to the central laboratory. The highest prevalence in people over 50 years old was 90.6% and the lowest incidence in the group under 9 years old was 9.4 percent, which were in agreement with the present study (30).

In recent experiments conducted on animals, the female animals that are negative for *Toxoplasma* serology are infected after having sex with a male animal infected with toxoplasma. Besides positive titration, the female animals have the ability of vertically transmitting this infection to their children, similar to human. Therefore, the transmission of parasites from mothers to fetuses that has also been observed before is confirmed (8, 9).

In the present study, there was no significant correlation between the frequency of anti-*Toxoplasma* IgG and variables such as job, vegetable washing, pet contact, abortion in women, and semi cooked meat consumption. Correspondingly, this is similar to a study, Modrek et al (24) on adult girls to indicate how raw vegetables are consumed, and is nonconsistent with the research conducted by Bittencourt (31) on seroepidemiology of toxoplasmosis in pregnant women (24, 31).

Conclusion

Considering the significant increase in the prevalence of anti-*Toxoplasma* IgG in postmarital women and its fix stay in virgin girls and multi partners' women, which is related to aging and number of children but it is not significant, so it can be concluded that the increased titer probably is due to sexual activity. Although this is not the only parasite transmission way, but it can be considered as a method of transmission. Awareness and education is necessary to prevent the transmission of sexually transmitted infections including *Toxoplasma* in different societies.

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Conflict of interest

The authors declare that there is no conflict of interest.

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