

Original Article

Seroprevalence of toxoplasma antibodies in women with normal pregnancies and history of abortions

Kulshrestha Simta¹, Solanki Aruna², Khatri PK³

¹ Sr. Demonstrator, ² Associate Professor & Head, ³ Assistant Professor

Department of Microbiology, Dr. S. N. Medical College, Jodhpur.

Abstract

Objectives: To study the seroprevalence of antitoxoplasma antibodies in normal non pregnant, normal pregnant and in women with history of abortions according to age, living conditions, history of association with animals, dietary habits, period of gestation and frequency of abortion whether sporadic or habitual. **Methods:** The study was conducted in the Department of Microbiology, attached to Dr. S. N. Medical College, Jodhpur in Rajasthan. It included 50 non pregnant (control group I) 50 normal pregnant (control group II) and 500 women (study group) having history of abortion. Standardized quantitative ELISA test was performed for Toxo IgG and Toxo IgM antibodies to *Toxoplasma gondii* (T. gondii) in sera of subjects included in the study. **Results:** Seroprevalence of toxoplasmosis was more in women with history of abortion. Overall seropositivity of Toxo IgM was maximum in the age group of 15-25 years (6.25%), whereas seroprevalence of Toxo IgM increased with age and was maximum (16.67%) in the age group 26-35 years in study group. There was increased prevalence of toxoplasma antibodies in both rural and urban population but percentage of Toxo IgM antibody was increased in normal pregnant (100%) and in study group females (28.57%) among rural population. There was increased prevalence of Toxo IgG and IgM antibodies in both control and study group females having association with animals such as cat and dogs. In the study group, in women having association with animals like cats and dogs, there was increased percentage of both Toxo IgG (38.10%) and Toxo IgM antibody (14.29%) as compared to those in normal pregnant control group - Toxo IgG (9.09%) and Toxo IgM (6.82%). Maximum prevalence of toxoplasma was also seen in study group women who were non vegetarians - Toxo IgG (50%) and Toxo IgM (14.29%). The prevalence in the study group was more in second trimester of pregnancy and with history of habitual abortion - IgG (28.29%) and Toxo IgM (18.75). **Conclusion:** It can be concluded that all subjects with bad obstetric history should be screened for toxoplasmosis and if found positive should be treated to prevent fetal losses.

Key words: toxoplasma, antibodies, pregnant, non pregnant, abortion

Introduction

The prevalence of toxoplasmosis in women with bad obstetric history is known to be significantly higher

than in normal women (Mahajan et al, 1976). Approximately 30% of the women of child bearing age in the United States have antibodies against *Toxoplasma gondii* (Dubey and Beattie, 1998) and remaining 70% of the women are at risk of acquiring *Toxoplasma gondii* infection during pregnancy. There is a 20%-50% probability that her fetus will be infected³. The seroprevalence in normal pregnant women on worldwide basis varies from 7-51.3% and in women with abnormal pregnancies and abortion, the seroprevalence

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Correspondence :

Dr. Aruna Solanki

18, Amarnath Building, Opposite M.G. Hospital

Jodhpur (Rajasthan)

Tel. 0291-2626299; Email : solanki.aruna@gamil.com

varies from 17.5 to 53.3%. Singh⁴ and Soliman et al⁵ found a significant difference of anti toxoplasma antibody titer in normal and complicated pregnancy cases. Kumar⁶ also observed statistical difference between IgG level in habitual abortion group as compared to sporadic abortion group or normal pregnancy group.

The maternal infection is often asymptomatic or it results in clinical disease that is not recognized. Antibody screening programs aimed at the diagnosis of T. gondii infection among pregnant women have therefore been introduced in several countries to control fetal loss and transplacental infection in children.

Not much is known about prevalence of toxoplasma antibodies in normal pregnant women and in patients with history of abortions in this western region of Rajasthan. Hence the present study was undertaken to find out the prevalence of anti toxoplasma antibodies in normal non pregnant, normal pregnant and in patients with history of abortions according to age, living conditions, history of association to animals and frequency of abortion.

Methods

The study was conducted in the Department of Microbiology attached to Dr. S. N. Medical College, Jodhpur in Rajasthan. The study included 50 non pregnant women (control group I), 50 pregnant otherwise healthy women (control group II) and 500 patients (study group) having history of abortion in the form of sporadic and habitual abortion. The control group II and study groups were selected after taking proper history from indoor and outdoor patients coming to Umaid Hospital attached to Dr. S. N. Medical College, Jodhpur. The control group I was selected from the staff and students of this institution. Blood sample from all the three groups were collected in sterile containers without anticoagulant. Sera were separated from these samples and stored at 2-8°C for a short period till analyzed.

Standardized quantitative ELISA test was performed for IgG and IgM antibodies to Toxoplasma gondii in the sera samples as per the technic described in the kit literature. The kits used were supplied by Sero Quest, North Miami (USA). Test results were read as index value for IgM. Index value less than one, were

Table 1. Anti toxoplasma IgG and IgM antibody levels of the subjects under study according to age in years.

Control I Non Pregnant group age in	Total		Toxo IgG Negative <30Iu/ml		Toxo-IgG Positive 30-200 IU/ml		Toxo IgM Negative Index <1.0		Toxo IgM Positive Index >1.0	
	No.	%	No.	%	No.	%	No.	%	No.	%
15-25 years	32	64	30	93.75	02	6.25	29	90.62	03	9.38
25-35 years	16	32	16	100	00	00	15	93.75	01	6.25
36-45 years	02	04	02	100	00	00	02	100	00	00
Total	50	100	48	96	02	04	46	92	04	08
Control II Pregnant group										
15-25 years	34	68	30	88.24	04	11.76	32	94.12	02	5.88
25-35 years	14	28	13	92.86	01	7.14	13	92.86	01	7.14
36-45 years	02	04	01	50	01	50	01	50	01	50
Total	50	100	44	88	06	12	46	92	04	08
Study group										
15-25 years	310	62	190	61.29	120	38.70	280	90.32	30	9.68
25-35 years	180	36	160	88.89	20	11.11	150	83.33	30	16.67
36-45 years	10	02	10	100	00	00	10	100	00	00
Total	500	100	360	72	140	28	440	88	60	12

considered negative and index value more than one were considered positive. Toxo IgG antibody titer less than 30 IU/ml were considered negative and titers between 30-200 IU/ml or above were considered positive, indicating exposure to toxoplasma in the recent

past or past exposure to infection which is still active. The test results read as index value for IgM and international unit/ml (IU/ml) for IgG antibodies of toxoplasma are traceable to WHO anti toxoplasma serum, 3rd International standard preparation 1994.

Table 2. Anti Toxoplasma IgG and IgM antibody levels of the subjects under study according to their living conditions.

Control I Non pregnant	Total		Toxo IgG Negative <30 IU/ml		Toxo IgG Positive 30-200 IU/ml		Toxo IgM Negative index <1.0		Toxo IgM Positive index >1.0	
	No	%	No	%	No	%	No	%	No	%
Urban	38	76	36	94.74	02	5.26	34	89.47	04	10.53
Rural	12	24	12	100	00	0	12	100	00	00
Total	50	100	48	96	02	04	46	92	04	08
Control II Pregnant group										
Urban	48	96	42	87.50	06	12.50	46	95.83	02	4.17
Rural	02	4	02	100	00	00	00	00	02	100
Total	50	100	44	88	06	12	46	92	04	08
Study group										
Urban	430	86	290	67	140	32.56	400	93.02	30	6.98
Rural	70	14	70	100	00	00	50	71.43	20	28.57
Total	500	100	360	72	140	28	450	90	50	10

Table 3. Anti toxoplasma IgG and IgM antibody levels of the subjects under study according to association to animals.

Control I Non pregnant	Total		Toxo IgG Negative <30 IU/ml		Toxo IgG Positive 30-200 IU/ml		Toxo IgM Negative index <1.0		Toxo IgM Positive index >1.0	
	No	%	No	%	No	%	No	%	No	%
Present	15	30	13	86.67	02	13.33	13	86.67	02	13.33
Absent	35	70	31	88.57	04	11.43	32	91.43	03	8.57
Total	50	100	44	88	06	12	47	94	03	06
Control – II Pregnant Group										
Present	44	88	40	90.91	04	9.09	41	93.18	03	6.82
Absent	06	12	06	100	00	0	05	83.33	01	16.67
Total	50	100	46	92	04	08	46	92	04	08
Study Group										
Present	210	42	130	61.90	80	38.10	180	85.71	30	14.29
Absent	290	58	230	79.31	60	20.69	270	93.10	20	6.90
Total	500	100	360	72	140	28	450	90	50	10

Table 4. Anti toxoplasma IgG and IgM antibody levels of the subject under study according their dietary habits.

Control I Non pregnant	Total		Toxo IgG Negative <30 IU/ml		Toxo IgG Positive 30-200 IU/ml		Toxo IgM Negative index <1.0		Toxo IgM Positive index >1.0	
	No	%	No	%	No	%	No	%	No	%
Vegetarians	34	68	32	94.12	02	5.88	31	91.18	03	8.82
Non Vegetarians	16	32	12	75	04	25	15	93.75	01	6.25
Total	50	100	44	88	06	12	46	92	04	08
Control-II Normal pregnant group										
Vegetarians	36	72	35	97.22	01	2.78	34	94.44	02	5.56
Non Vegetarians	14	28	13	92.86	01	7.14	12	85.71	02	14.29
Total	50	100	48	96	02	04	46	92	04	08
Study group										
Vegetarians	360	72	300	83.33	60	16.67	330	91.67	30	8.33
Non Vegetarians	140	28	70	50	70	50	120	85.71	20	14.29
Total	500	100	370	74	130	26	450	90	50	10

Table 5. Anti toxoplasma IgG and IgM antibody levels of the subjects according to period of gestation.

Control I Non pregnant	Total		Toxo IgG Negative <30 IU/ml		Toxo IgG Positive 30-200 IU/ml		Toxo IgM Negative index <1.0		Toxo IgM Positive index >1.0	
	No	%	No	%	No	%	No	%	No	%
1-3 months	08	16	07	87.50	01	12.50	07	87.50	01	12.50
4-6 months	26	52	25	96.15	01	3.85	24	92.31	02	7.69
> 6 months	16	32	16	100	00	00	15	93.75	01	6.25
Total	50	100	48	96	02	04	46	92	04	08
Study group										
1-3 months	153	30.60	94	61.44	59	38.56	138	90.20	15	9.80
4-6 months	184	36.80	133	72.28	51	27.72	154	83.70	30	16.30
>6 months	163	32.60	124	76.07	39	23.93	144	88.34	19	11.66
Total	500	100	351	70	149	29	436	87.20	64	12.80

Table 6. Anti toxoplasma IgG and IgM antibody levels of the subjects under study according to frequency of abortion.

Frequency of Abortion	Total		Toxo IgG Negative <30 IU/ml		Toxo IgG Positive 30-200 IU/ml		Toxo IgM Negative index <1.0		Toxo IgM Positive index >1.0	
	No	%	No	%	No	%	No	%	No	%
Control II Normal pregnant group	50	100	44	88	06	12	46	92	04	08
Study group										
Sporadic abortion	249	49.80	195	78.31	54	21.69	216	86.75	33	13.25
Habitual abortion	251	50.20	180	71.71	71	28.29	204	81.27	47	18.73
Total	500	100	375	75	125	25	420	84	80	16

Results

It has been observed that 12% of the women in control group II (normal pregnant) and 4% of the women in control group I (Non pregnant) had positive Toxo IgG levels. Out of the 50 controls (group I & II) 46 (92%) had negative Toxo IgM and 8% had positive Toxo IgM levels. Similarly 28% of the patients with history of abortions had positive Toxo IgG levels and 12% of them had positive Toxo IgM levels. However, overall increased positive rate for Toxo IgG antibodies was seen in the age group 15-25 years followed by the age group 26-35 years in group II and the study group. Increased positive rate for Toxo IgM was seen in the age group 26-35 years (Table 1).

Table 2 revealed that amongst the control group I and II, subjects residing in urban area (5.26% of non pregnant females and 12.50% of normal pregnant females) had raised Toxo IgG levels, whereas 32.56% of the study group women residing in the urban area had positive Toxo IgG levels. None of the rural residents (non pregnant, normal pregnant and women with history of abortions) had increased Toxo IgG levels.

It has been observed that maximum seropositivity (100%) for Toxo IgM was seen in rural residents of group II (normal pregnant) followed by 28.57% of the rural residents of females with history of abortions (Table 2).

Table 3 shows that percentage of Toxo IgG and IgM levels were raised in cases having close association to animals like cats and dogs in comparison to cases without history of close association to animals, except control group II (normal pregnant 16.67%) where increased percentage of IgM levels were seen in cases without history of close association to animals.

The comparison of Toxo IgG antibodies of non vegetarian healthy pregnant and non pregnant control group with that of study group (non vegetarians) showed that 50% cases with history of abortion were positive for Toxo IgG antibodies and 7.14% of the normal pregnant and 25% of non pregnant, non vegetarian control group were positive for Toxo IgG antibodies. The percentage of Toxo IgM antibodies in non vegetarian cases of the study group was equal to that of healthy pregnant group (14.29%).

Whereas, Toxo IgM percentage in the non vegetarian, on pregnant control group was only 6.25%, which is very low compared to that in the pregnant control (group II) and the study group. Toxo IgM percentage in vegetarians varied from 5.55 to 8.83% (Table 4).

In pregnant control group, out of 52% cases with period of gestation of 4-6 months, 3.85% cases showed positive Toxo IgG and 7.69% showed Toxo IgM antibody level. In the study group, 36.80% of the patients belonged to 4-6 months of gestational period and seropositivity for Toxo IgG and Toxo IgM antibody were 27.72% and 16.30% respectively. Seropositivity in patients with history of abortions was greatly increased both for Toxo IgG and Toxo IgM antibodies (29.80% and 12.80% respectively) as compared to that in the pregnant control group (4% and 8% respectively) (Table 5).

Table 6 showed that seroprevalence of Toxo IgG was 29.29% and 21.69% in the study group with habitual abortions and sporadic abortions respectively as a compared to only 12% in the healthy pregnant control group. The percentage of Toxo IgM was maximum (18.75%) in patients with habitual abortions followed by 13.25% in patients with sporadic abortions and 8% in normal healthy control group.

Discussion

The study reveals that toxoplasmosis is not an uncommon disease in this region of Western Rajasthan. The prevalence of toxoplasmosis according to age has been studied by many workers and it has been observed that human infection begins at an early age and it increases with age⁸⁻¹⁰.

In the present study, overall seropositivity for Toxo IgG was maximum in the age group of 15-25 years followed by 26-35 years. However, seroprevalence for Toxo IgM increased with age, and was maximum (16.67%) in the age group 26-35 years, in patients with a history of abortion (Table 1). This increased level of Toxo IgG and IgM antibody level in the age group of 15-30 years may be due to extra care taken in selection of the cases from reproductive age group.

Increased percentage of Toxo IgG antibodies was seen in non pregnant (5.26%), normal pregnant (12.50%) and study group (32.56%) in urban population as compared to that in rural population. The percentage of Toxo IgM was (100%) in normal

pregnant women of rural population as compared to urban population (Table 2). Therefore it is evident that increased *Toxoplasma* antibodies were found in both urban and rural population, but the percentage of Toxo IgM antibody was increased in rural than in urban population. IT may be due to environmental factors, i.e. (extremes of summers and winters and very little rainfall may have little influence on the survival of oocyst) and poor socio economic conditions of the subjects which may be responsible for high prevalence of this infection. The study conducted by Kawashima et al¹⁰ indicated that there is higher seropositivity in rural than urban settings.

The association of toxoplasma antibodies in women with a history of keeping animals (cats at home) has been studied by many workers^{9,11}. In the present study, there was increased prevalence of toxoplasma antibodies, both Toxo IgG and IgM, in non pregnant, normal pregnant and in women with history of abortion having association with animals like cat, dogs, etc. (Table 3). These findings correlate with the study of Al-Hamdani⁹. These findings may be due to the fact that oocysts are excreted in feces of cat, and contamination of food, fingers or drink with cats feces may result in human infection¹². Earthworms, flies or cockroaches may serve as mechanical vectors of oocyst. Rarely infection may occur through consumption of water contaminated with cat feces containing oocyst¹³. This study also indicated that in the study group, in women with history of animal association, there was increased percentage of both Toxo IgG antibodies (38.10%) and IgM antibodies (14.29%), when compared with that in normal pregnant group having history of animal association (Toxo IgG 9.09% and Toxo IgM 6.82%) (Table 3).

The study conducted by McAuley¹⁴ showed that 28 out of the 45 women who had delivered congenitally infected children had handled or consumed under cooked meat during gestation. In the present study, overall seroprevalence of Toxo IgG was raised in non vegetarian women of all the three groups viz. non pregnant (25%), normal pregnant (7.14%) and in women with history of abortion (50%). The Toxo IgM was 14.29% in both vegetarian normal pregnant group and the study group women. The relatively high prevalence of Toxo IgG (50%) in study group women may be due to the fact that transmission of toxoplasma infection can also occur as a result of ingestion of under cooked meat harboring tissue cysts. Thus all these observations suggest that socio economic conditions, poor hygiene, along with improper dietary habits, illiteracy, for living

conditions and environmental factors influence the prevalence of toxoplasmosis.

It has also been observed that the prevalence rate of Toxo IgG in the healthy non pregnant women was 12% as compared to only 4% in the healthy pregnant group (Table 4). This could be because of the extra care taken in the selection of healthy pregnant females for this study. It may also be due to the immuno suppressive effect of pregnancy on immune system resulting in suppressed antibody formation.

The study group showed presence of both Toxo IgG and IgM antibodies in all the three trimesters. However Toxo IgM antibody levels were relatively high (16.30%) in the second trimester and IgG levels showed maximum prevalence (38.56%) in the first trimester (Table 5). Thus the study indicated that there is no correlation between these two types of antibodies with the duration of pregnancy.

Table 6 indicated an increased percentage of Toxo IgG (28.29%) and Toxo IgM antibodies (18.73%) in cases of habitual abortion as compared to those in cases of sporadic abortion (IgG 21.69%; IgM 13.25%). In normal pregnancies prevalence of Toxo IgG and IgM was 12% and 8% respectively. The seroprevalence of toxoplasma antibodies in normal pregnancies and in abortion group has been studied by many workers^{4,6,9}. Our findings correlated with the findings of Soliman et al⁵ and Kumar⁶. The experience of several obstetricians has proved beyond doubt that pregnancy outcome is definitely better in most of the women who took regular and full treatment and treated women also delivered babies without any signs of toxoplasmosis. It has also been observed that in untreated seropositive women, incidents of abortions were greatly increased. Hence after getting the test results from the microbiology department, these seropositive women were treated with spiramycin and pyrimethamine plus sulphadiazine drug schedule. Drug of choice was spiramycin in the doses of 6-9 IU/ml per day in 2-4 divided doses for three weeks, pyrimethamine 25-50 mgm. per day with 2-3 gm. of sulphadiazine, and folic acid 5 mg. per day for three weeks in the late second and third trimester.

Thus it can be concluded that toxoplasmosis is one of the important causes of abortion in this region. As such it is recommended that all such subjects be screened for toxoplasmosis and if found positive, should be treated to prevent further transmission of organisms to fetus and prevent abortion and fetal loss. Thus for

proper assessment of the presence and absence of toxoplasmosis and the state of disease process, it is recommended to assay both Toxo IgG and Toxo IgM antibody in pregnant women and in women with history of abortion.

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