

INTERNATIONAL JOURNAL OF SCIENCE AND NATURE

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SEROPREVALANCE OF TOXOPLASMIC CHORIORETINITIS IN BAGHDAD PROVINCE

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ABSTRACT

The study was carried out to investigate the prevalence of toxoplasmic chorioretinitis in Baghdad province. Samples of serum belonged to 102 patients (40 males and 62 females) of different age with ocular lesions were collected during the period between October 2010 and May2012 from different areas of Baghdad province. ELISA test was performed to demonstrate the rates of antitoxo IgG and IgM antibodies, using vercill kit. The results revealed that that the infected percentage was 42(41.17%), forty out of them (95.23%) had positive anti toxo IgG antibodies and only2 (4.76%) had positive anti IgM antibodies. Significant differences were recorded regarding the gender. The positive cases were 15 (35.71%) out of 42 positive cases of males, whereas the cases in women were 27 (43%). All the males had positive anti toxoIgG antibodies, while in case of females, 25(62.5%) had positive anti toxoIgG antibodies and 2case (100%) had positive IgM at age over 35 years old. The results display that 17(70.58 %) women were aborted and 10 (37.03%) non aborted. Most women are employed19 (70.37 %) the rest 8 (28.62%) are housewives. The rate of abortion was higher within an employed women 12(70.58%) compared to housewives 5(29.41%). Women in urban and rural areas were vulnerable to infection. Abortion may due to recent exposure to infection or due to reactivation past infection. The effect of age on infection rate was not significant. All age groups in either children or adults showing rates of infection with toxoplasmic chorioretinitis (100%). Regarding the type of infection, results obtained that the lesion of chorioretinitis due *Toxoplasma gondii* in one eye 30 (71.42 %) differed significantly compared to both eyes12 (28.57 %).

KEY WORDS: Toxoplasmic chorioretinitis, Serological diagnosis

INTRODUCTION

Toxoplasma gondii is an a coccidian parasite of the cat, it has been found to be worldwide distribution, most common in warm moist can infect almost any warm blood animals or birds and humans (1,2,3). Toxoplasmosis may be congenitally acquired in utero during the second or third trimester of fetal development usually from a symptomless mother (4) or acquired post natal by ingesting tissue cysts in under cooked or uncooked meat or by food and water contaminated with oocysts from infected cat feces. Infection is usually asymptomatic and self limited condition can go unrecognized in 80% to 90% of adult and children with acquired infection (5). When toxoplasmosis symptomatic the most commonly organ attacked by parasite are the lymph nodes, brain, eyes, lung and frequently involves the central nervous system which is characterized by aneurologic sequelaeintra cerebral calcification, seizures, mental retardation, chorioretinitis and blindness. Chorioretinitis are late sequelae of congenital toxoplasmosis. The most seriously affected human cases are those of the newborn, some infants develop ocular lesions as children and young adults, the pathologic process causes chorioretinitisat time with hydrocephalus preumonitisandmyocorditis (6). In persons whose immune system are intact, disease acquired later in life and retino choroiditis may be manifestation toxoplasmic chorioretinitis can occur as result of are cently acute acquire defection or reactivation. Typical findings of toxoplasmic chorioretinitis are white focal

lesions with an overlying and intense vitreous inflammatory reaction and the retinal detachment associated with toxoplasmic. Retinal detachment associated with toxoplasmic retinochoroiditis is rare however it represents a serious complication which is lead to blindness (7). A toxoplasmic chorioretinitis occurring by *Toxoplasma gondii* infection may be common throughout Iraqi people and toxoplasmic serological profile is useful to early diagnosis and treatment this entity.

MATERIALSANDMETHODS

Bloodsample: During the period between October 2010 and May 2012, a total of 102 blood samples collected from patients comprising 40 males and 62 females of different age, from urban and rural areas of Baghdad province admitted from Ibn Al-Hitham Hospital of Opthalmic to the Central Public Health Laboratory, who had ocular lesions suspected with toxoplasmosis. Serum samples were prepared and stored at 20°c until used.

Serologicaltest: Serum samples examined using Vircell kit for specificant toxo IgG and IgM antibodies, received from (Vircell Company) Spain. The Batchnumber of IgG is 12ETOX G102 and of IgM is 12ETOXC103. A qualitative ELISA test standardized and performed to estimate the immunological response of these individual by analyze 102 sera samples, positive and negative serum, regarding the gender, age and socio inform.

IgG kit preparation: Wells coated with *T. gondii* antigen strain RHI (Atcc50174).1-Add100Ul, sample diluents to all wells, add 5UL. of each sample, negative, positive control and cut-off (in duplicate) into corresponding wells. 2-cover with a sealing sheet and incubate at37°C for 45 min. 3-Remove the sheet, wash plate 5 times, drain any remaining liquid.4-Add 100UL, of IgG conjugate solution to all wells. 5- Cover with a sealing sheet and incubate at 7°C for 30min. 6-Remove the sheet, wash plate 5times, drain any remaining liquid.7-Add 100UL of substrate solution into all wells.8- Cover with a sealing sheet and incubate at room temperature for 205 min.9-Add 50UL.of stopping solution (0.5M H2SO4) to all wells10-Read with an spectrophotometer at 450/620 nm within 1h.of stopping.

Interpretation of results: Antibody index = (sample OD/cut off serum mean OD) X10. The sample is negative if the index (<9). The sample is equivocal if the index (9-11). The sample is positive if the index (>11).

IgM kit preparation: Wells coated with anti IgM antibodies. Conjugate solution must be prepared in advance, at least 1hbeforeused.Add3 ml. of reconstitution solution to one vial of lyophilized conjugate, let stand for 1 min to allow rehydration and mix thoroughly by vortex. The rest steps performed for IgM preparation the same as in IgG antibody prepared follow the instruction of manufacture. Interpretation of results is the same for IgG antibodies.

Statistical Analysis: Chi-square test (x^2) was used to compare ratio of IgG and IgM antibodies and infection rates. The differences were considered statistically significant when the P value less than 0.05 (8).

RESULTS AND DISCUSSION

Diagnosis of acute toxoplasmosis is usually accomplished serologically because symptoms are very non specific (9). Acutely IgM antibodies appear for several months followed by IgG antibodies that persist provide lifelong protection (10). By using enzyme immune assay a quantitative test we have conducted a detailed research on the toxoplasmic chorioretinit is using vircell kitto measure the ratio of anti-toxoplasma IgG and IgM antibodies in the sera of patients with ocular lesion(11,12). The Seroprevalance of *T. gondii* infection has been recorded to be high. Out of 102 serum samples examined, 42(41.17%) had positive anti toxo antibodies. Forty (95.23%) had positive anti toxo IgG antibodies and only2 (4.76%) samples had positive antitoxoIgM antibodies at age over than 35 years old. In this research, the total rate of infection was higher than those reported comparable to the in Baghdad province (13) and in Basra province (14), the hot and arid climatic condition are associated with low prevalence of the infection in Basra. Seroprevalance rate also was higher than in some other studies performed in other countries (15, 16,17,18) which attributed to climate conditions and urban life. Results revealed that rate of infection in males 15(35.71%)was differed significantly (P<0.05) compared to females 7(64.29%). All the infected males had positive anti toxoIgG antibodies, the ratio between(16-34), while in case of females25case (92.59%) had positive IgG antibodies and the ratio between 11.8-66 and 2patients(28.5%) had positive IgM antibodies, the ratio was between 19.5-22(Table1).

TABLE1: The effect of gender on seroprevalance of Toxoplasmic chorioretinitis

Serum samples	No	+ve no	%	IgG+ve no	Range	%	Mean± SD	IgM +ve no	%	Range	Mean±SD
Males	40	15*	35.71	15*	16-34	100	22.79±1.41	_			
Females	62	27*	64.28	25*	11.8-66	92.59	22.65 ± 2.53	<u>2</u> *	$\frac{-}{28.5}$	19.5-22	$\overline{4.30\pm0.95}$
Total	102	42	41.17	40*	11.8-66	95.23	26.72±4.29	2*	4. 76	19.5-22	4.44 ± 0.95
											*P < 0.05

1 .0.

TABLE 2: The rate of infection in aborted and non aborted employed and housewives

Company gomentos			Employe	ed	Housewives		
Serum samples	+ve no	(%)	ve no+	(%)	ve no+	(%)	
Aborted	17	62.96*	*12	70.58	5*	29.41	
Non aborted	10	37.03*	7	70	3	30	
Total	27	64.29*	19	70.37	8	29.62	

*P < 0.05

Table (2) display that the rate of aborted womenwas17 (62.96 %) and 10 (37.03%) non aborted women, most women in this research areemployed19 (70.37 %) the rest 8 (29.67%) are house wives, the results revealed that the rate of abortion in employed women was higher12 (70.58%)than housewives5(29.41%). Women from both urban and rural areas were vulnerable to infection. Abortion may due reactivation of a previous infection rather than acquiring new infection from their cat or other sources. In our research, women infection more than men as a result to their duties in the house, exposure to infection may occur when handling with meats or cleaning gardening were outdoor cats are a round, the rate of

infection in women was higher than the rates(13.7%), (11%) in Iraq (13, 14) and (15.33%) of (13,14,4)in India. Toxoplasmosis Seroprevalance depend on the location and the age of the population(19). The results illustrated in the table (3) revealed that no significant differences regarding the age, toxoplasmosis display in a high rates in all age groups, among the (children) (>10) and in age group between(11_20) years, all patients had(100%) positive anti toxoIgG antibodies. About the age group of (21_30) years, out of 9(60%) only onecase (11.1%) had positive antitoxo IgM antibodies. Within the age over 30 and 40< years it reach to13 (92.85%) and11 (100 %) respectively. The ingestion of oocyst from an environment heavily

contaminated with ooctys from cats feces or drinking un filtered water can acquired without doubt in adult and children in urban and rural areas in Baghdad province due to poor hygiene measurement in most areas and the infection at the retinal neurons may occur years after the original acute infection has passed (6). These findings were corresponding well with the observation of(13).(14) demonstrated that the rate of incidence increase with age 34 years old reaches(17.8%)which is lowest than our result, while(16)recorded (3%) in women at age of 35-45years those have cats in their houses. The lowest (24.3%)rate being in the northern parts in India and the highest in the south(4) These variation in the rates may due to number examined. Toxoplasmic chorioretinitis can occur as result of infection or reactivation. Persistence of

the encysted organism may lead to reactivation of infection which can cause severe symptoms of toxoplasmosis in later age, especially in person with severely weakened immune system (7, 20, 21). In this research, the results revealed significant differences regarding the type of infection. The infection rates of patients with ocular lesions in both eyes were 12(28.57%) and 30(71.42%) in one eye (14 in the left eye and 16 in the right one). Significant differences were recorded between IgG and IgM antibodies ratio. All patients (30) infected with one eye had (100%) anti toxoIgG antibodies. While of the 12(28.57%) which represent infection in both eyes 10(83.3%) of patients had anti toxoIgG antibodies and 2(16.66%) patients had anti toxoIgM antibodies (Table4).

TABLE 3: Seroprevalance of Toxoplasmic chorioretinitis regarding the age

Age (years)	No of case	+ve No	%	No of IgG	%	Mean+SE of IgG	No. of IgM	%	Mean+ SE of IgM
>10	10	5	50	5	100	22.00±2.51	- 0		
11-20	12	3	25	3	100	16.35 ± 2.26	_	_	_
21-30	15	9	60	8	88.88	22.00±2.51	1	$\bar{1}1.1$	$\frac{-}{5.13\pm2.16}$
31-40	35	14	40	13	92.85	24.00 ± 3.03	1	7.14	4.07±1.26
41<	30	11	36.66	11	100	21.39±1.89	_	_	_
Total No	102	42	41.17	40	95.23	26.72±4.29	2	4.76	4.44±0.95

Non-significant

TABLE 4: The type of infection with toxoplasmic chorioretinitis

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Lesions	+ve No	%	IgG +ve No	%	Ranged	Mean± SD	IgM +ve no	%	Ranged	Mean± SD
One eye	30	71.42	30*	100	11.8-66	21.40±1.52	_	_	_	_
Both eyes Total	12 42	28.57 41.17	10* 40	83.33 95.23	12.8-34 11.8-66	24.00±3.03 26.72±4.29	2 2	16.66 4.76	19.5-22 19.5-22	3.96±0.94 4.44±0.95
										D<0.05*

P<0.05*

The differences of these results with the results of (6) whichshow5 (23.8%) had lesions in both eyes and 16 (76.1%) in one eye, could attributed to differences of the number of patients in this research, and the ratio of immunoglobulin (IgG) toxoplasmic antibodies in serum ranged between 6 and 56.(22) demonstrated that IgG-class anti-toxoplasmic antibodies produced locally within the eye in cases of toxoplasmic chorioretinitis (23). The continuously exposure to infection due to different causes lead to persist IgG in high level like booster dose cause latent immunity. Also inhibition in the level of IgM upon competent inhibition of IgG or upon blocking effect of IgG when found in high level (23,24). The results concluded that ocular infection involvement due to T.gondii infection is commonly in the people of Baghdad province. Results indicate that the ratios of IgG-class antitoxoplasmic antibodies were increased in serum of females and males patients.

The determination of the antitoxo G and M antibodies can offer a valuable aid to make specific diagnosis of ocular toxoplasmic chorioretinitis and treatment earlier to avoid a serious complication, laterretinal detachment.

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