The Use of Microparticles Enzyme Immunoassay in a Seroepidemiological study of Toxoplasma gondii antibodies in the Eastern Region of Saudi Arabia

Abdul Rehman Al-Qurashi
Departments of Microbiology,
College of Medicine, King Faisal University,
Dammam, Saudi Arabia

ABSTRACT:

Toxoplasma gondii is an obligate intracellular protozoan parasite with a worldwide distribution. It is particularly common in warm and moist climates. All the published work about Toxoplasma in Saudi Arabia used the conventional methods for detection of toxoplasma-specific antibodies (either haemagglutination, latex agglutination, immunofuorescent or ELISA). None of these studies used the more recent microparticle enzyme immunoassay (MEIA) for analysis of antibody responses to Toxoplasma. In this study, we used the Toxoplasma IgG MEIA to analyze sera from 1464 individuals from different parts of the Eastern Region of Saudi Arabia. Out of 1464 sera tested for IgG 1100 (75.14%) were found to have negative values less than 2.94 IU/ mL while 364 (24.86%) were recorded with positive values of equal to or more than 2.94 IU/mL. To our knowledge, this is the first report of the use of MEIA in a population based seroepidemiological study of toxoplasma antibodies in Saudi Arabia.

In conclusion, we found the MEIA very practical for our study and we recommend this technique for use in large seroepidemiological studies with large sample size.

Key Words: Microparticle Enzyme Immunoassay, Toxoplasma gondii, Eastern Region of Saudi Arabia.

INTRODUCTION:

Toxoplasma gondii is an obligate intracellular protozoan parasite with a worldwide distribution. It is particularly common in warm and moist climates. Domestic cats are the definitive hosts of the parasite. Man, livestock and even rodents may act as intermediate hosts for the parasite. Human infection may occur via contact with faeces of infected cats, ingestion of tissue cysts (bradyzoites) in undercooked or raw livestock's meat (Frejz and Sever, 1991, Heynmen and Goldsmith, 1992) or by blood transfusion.

Most of the Toxoplasma infections in adult males or females are either asymptomatic or benign. The infection is however, severe or fatal in neonates, in patients with acquired immunodeficiency syndrome (AIDS) and in immunocompromised patients.

Intrauterine (congenital) transmission may occur. Foetal involvement may result in hydrocephalus, intracranial calcification and mental retardation in infants born to infected mothers.

The prevalence of seropositivity of Toxoplasma ranges from 7.5 – 95% in different parts of the world: 7.5% in Scotland (Jackson and Hutchenson, 1987), 50% in USA (Stagno, 1980), 54.0% in Kenya (Grieffin and William, 1983), 47% in Nigeria (Onadelko et al. 1992), 37% in Jordan (Morsy and Michael, 1980), and 95.5% in Kuwait (Behbehani and Al-Karmi, 1980).

Few studies have been published on the pravalance of toxoplasma antibodies in Saudi Arabia (Amin and Morsi, 1997; Al-Amari, 1994; Sarwat et al 1993; Al-Harthi et al, 1988; Abbas et al, 1986). All the published work about Toxoplasma in Saudi Arabia used the conventional methods for detection of toxoplasma-specific antibodies (either haemagglutination, latex agglutination, immunofuorescent or ELISA). None of these studies used the more recent microparticle enzyme immunoassay (MEIA) for analysis of antibody responses to Toxoplasma.

The diagnostic technique (MEIA) used in the present study is rather a new technique and has been documented by other research workers (Petithory et al 1996). The technique was evaluated at 15 clinical sites in

Europe and USA for toxoplasmosis and other diseases. However, no evaluation was done for this method in a seroepidemiological study in the Kingdom of Saudi Arabia. A high sensitivity (97.0%) was recorded and a high specificity (99.8%) as well (Lyasu et al. 1995).

The MEIA is a fully automated system for measuring specific antibody by interaction with antigen-coated particles. MEIA uses the IMX immunoassay analyzer (Abott, USA). The IgG MEIA results are expressed in International Units (IU) of IgG antibody interpolated from a six point calibration curve covering the range from 0 to 300 IU/ml. Reproducible results were obtained from a calibration curve stored in the instrument for at least one month. The qualitative IgM MEIA expresses results as an index using a single calibrator included in each run. Previous studies have shown that the Toxoplasma IgG MEIA and Toxoplasma IgM MEIA were in 98% and 97% agreement, respectively, with the reference assays used (Sarwat et al 1993). The reference assay used were Haemagglutinate, Immunofluorescent and slide agglutination.

In this study, we used the Toxoplasma IgG MEIA to analyze sera from 1464 individuals from different parts of the Eastern Region of Saudi Arabia.

MATERIALS AND METHODS

The epidemiological study was population based (not hospital based thus differing from the previous studies of toxoplasmosis in Saudi Arabia). A cross sectional survey was conducted to estimate the seroprevalence of human toxoplasmosis in Eastern Saudi Arabia. Five areas were selected at random: three rural, Al Jisha, Al-Qurain, Al-Nereiyah, and two urban, Al-Mubaraz and Al-Khafgy (first stage in sampling). Stratified sampling was used to select at random from each of the five areas (according to population size) one or several segments of houses (second stage in sampling). A total of 1464 sera were collected in the study.

Sera were stored at -20^0C for further analysis by the Microparticle Enzyme Immunoassay (MEIA) for IgG and IgM (Petithory et al 1996). In this test IMX Toxo IgG assay data are expressed in International Units (IU) of IgG.

RESULTS:

Sera from 1464 individuals from the Eastern Region of Saudi Arabia were analyzed for the presence of IgG antibodies specific for T. gondii using MEIA (IMX Toxo IgG). Data presented represents IgG antibodies levels expressed in IU/ml. IMX Toxo IgG assay results of less than 2.94 IU/mL were considered negative for IgG antibody to T. gondii. Results of greater than or equal to 2.94 IU/mL were considered positive for IgG antibody to T. gondii and may indicate past inactive infection according to the manufacturer's recommendations.

The results for distribution of IgG antibodies are given in Table 1 and Fig. 1. The range of values was 0.1 – 300 IU/mL. Out of 1464 sera tested for IgG 1100 (75.14%) were found to have negative values less than 2.94 IU/ mL while 364 (24.86%) were recorded with positive values of equal to or more than 2.94 IU/mL.

DISCUSSIONS:

Of the negative sera, 86.1% (948) gave values of <1 IU/ml Toxoplasma IgG antibodies. These individuals most probably did not come in contact with the T. gondii in their life.

63.7% (232) of the positive sera gave values between 6 and 39 IU/ml IgG antibodies. These represent the majority of sera of the positive individuals. This indicates exposure to the parasite antigen. This could be as a result of past toxoplasmosis or past subclinical asymptomatic infection or current infection.

Of the positive sera, 3.3% (12) showed values ≥ 300 IU/ml IgG Toxoplasma antibodies. These 12 patients shows no symptoms or sign of toxoplasmosis. These very high values may represent latent infection, current infection or a past infection in an individual who is a high responder. This high responder status is genetically determined and is under the influence of the HLA genes and the IR genes (Roits, 1997).

To our knowledge, this is the first report of the use of MEIA in a population based seroepidemiological study of toxoplasma antibodies in Saudi Arabia. The MEIA offers the advantage of speed (Twenty four sera can be completely processed in about 35 minutes) over the other

conventional techniques, specially for analysis of large number of samples. MEIA is also objective, quantitative and can be used to measure IgG (inactive disease) as well as IgM (active disease). The disadvantage of the MEIA is the capital needed to bye the IMX analyzer.

In conclusion, we found the MEIA very practical for our study and we recommend this technique for use in large seroepidemiological studies with large sample size.

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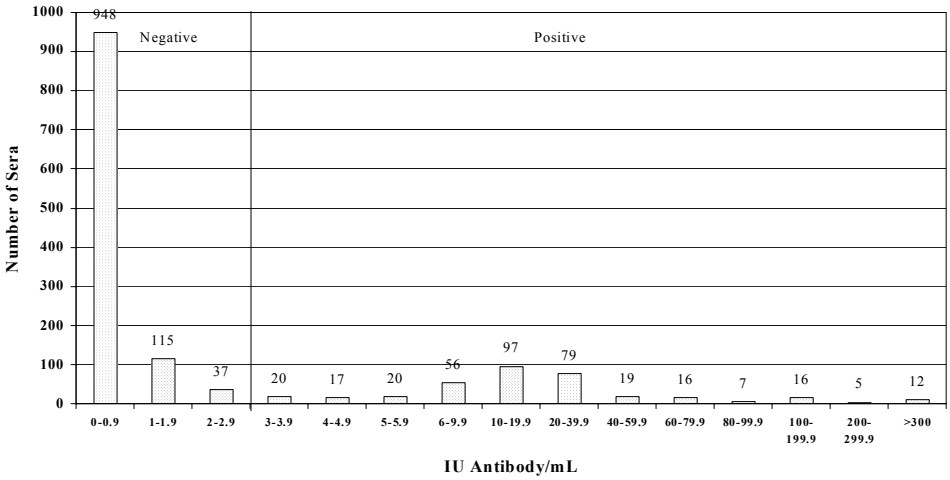
Table (1) : Overall Distribution of 1gG in Human sera examined in five areas of study in the Eastern Region

	Iu Antibody/mL	Number of Sera	% of Total
NEGATIVE	0.0 - 0.9	948	64.47
	1.0 - 1.9	115	7.85
	2.0 - 2.9	37	2.53
POSITIVE	3.0 - 3.9	20	1.37
	4.0 - 4.9	17	1.16
	5.0 - 5.9	20	1.37
	6.0 - 9.9	56	3.82
	10.0 - 19.9	97	6.62
	20.0 - 39.9	79	5.39
	40.0 - 59.9	19	1.29
	60.0 - 79.9	16	1.09
	80.0 - 99.9	07	0.48
	100 - 199.9	16	1.09
	200 - 299.9	05	0.34
	≥ 300 -	12	0.82

IgG Value < 2.94 Negative
≥ 2.94 Positive

Fig. 1: Overall distribution of IgG levels in human sera examined in five areas of study in the Eastern Region of Saudi Arabia



REFERENCES:

1) Abbas, S.A., Basalamah, A., Serebour, F. and Alfonso, M. (1986). The prevalence of Toxoplasma gondii antibodies in Saudi women and the outcome of congenital infection among Newborns in Saudi Arabia. Saudi Medic J, 7: 346-354.

2) Al-Amari, O.M. (1994). Prevalence of antibodies to Toxoplasma gondii among blood donors in Abha, Asir region, South Western Arabia. J Egypt Pub Heal Assoc, 69: 77-88.

3) Al-Harthi, S., Rehman, N., Bakir, T. and Al-Nozha, M. (1988). Cerebral Toxoplasmosis in the Acquired Immunodeficiency syndrome. Case Report and Review of Literature. Ann Saud Medic, 8: 292-296.

4) Amin AM and Morsy TA. (1997). Anti-toxoplasma antibodies in butchers and slaughtered sheep and goats in Jeddah Municipal abattoir, Saudi Arabia. J Egypt Soc Parasitol, 27:913-8

5) Behbehani, K. and Al-Karmi, T. (1980). Epidemiology of Toxoplasmosis in Kuwait. I. Detection of antibodies to T.gondii and percentage distribution among inhabitants. Trans Roy Societ Trop Medic Hyg, 74: 209-212.

6) Frejj, B.J., and Sever, J.L. 1991. Toxoplasmosis. Paed Rev, 72: 227-236.

7) Griffin, L., Williams, K.A.B. (1983). Serological and parasitological survey of blood donors in Kenya for toxoplasmosis. Trans Roy Societ Trop Medic Hyg, 6: 763-766.

8) Heyneman, D. and Goldsmith, R. (1992). Tropical Medicine and Parasitology, Prentice Hall International, 942 pp.

9) Jackson, M.H. and Hutchinson, W.M. (1987). A seroepidemiological survey of toxoplasmosis in Scotland and England. Ann Trop Medic Parasitol, 81: 395-365.

10) Lyasu, V., Robert, A., Schaefer, L. and Maciozek, J. (1995). Multicenter Evaluation of a new Commercial assay for detection of immunoglobulin M antibodies to Toxoplasma gondii, Multicenter Study Group. Europ J Clinic Microbiol Infect Diseas, 14: 487-493.

11) Morsy, T.T. and Michael, S.A. (1980). Toxoplasmosis in Jordan. J Egypt Societ Parasitol, 10: 457-470.

12) Onadelko, M.O., Joynson, D. H. and Payone, R.A. (1992). The prevalence of Toxoplasma infection among pregnant women in Ibadan, Nigeria. J Trop Medic Hyg, 95: 143-145.

13) Petithory, J.C., Reiter-Owona, I., Berthelot, F., Milgram, M., De Loye, J., and Pelersen, E. (1996). Performance of European laboratories testing serum samples for Toxoplasma gondii. Europ J Clinic Microbiol Infect Diseas, 15: 45-49.

14) Roit I. (1997). Essential Immunology, 10th Edition, Blackwell Science Press.

15) Safford JW, Abbott GG, Craine MC and MacDonald RG (1993). Automated microparticle enzyme immunoassays for IgG and IgM antibodies to Toxoplasma gondii. J Clin Pathol, 44:238-42

16) Sarwat MA, Ahmed AB, Zamzami OM, Fawzy AF and Morsy TA (1993). Toxoplasma gondii in Saudi blood donors a serological study using three tests. J Egypt Soc Parasitol, 23:751-7.

17) Stagno, S. (1980). Congenital toxoplasmosis. Amer J Dis Chil, 134: 635-637.

إستعمال طريقة الميا في دراسة وبائية عن طفيل التوكسوبلازما في المنطقة الشرقية للمملكة العربية السعودية

عبدالرحمن محمد القرشي

قسم الأحياء الدقيقة، كلية الطب، جامعة الملك فيصل
الدمام - المملكة العربية السعودية

الملخص :

يعيش طفيل التوكسيابلازما داخل الخلايا وهو ذو إنتشار عالمي، حيث تزيد نسبة الإصابة به في المناطق الحارة. كل البحوث المنشورة عن التوكسيابلازما في المملكة العربية السعودية استعملت الطرق القديمة للفحص. في هذا البحث ، تم استعمال طريقة الميا (MEIA) في الفحص على ١٤٦٤ عينة مجموعة من المنطقة بالمملكة العربية السعودية. نتيجة الفحص كانت كالتالي : ١١٠٠ (٧٥,١٤%) سلبية و ٣٦٤ (٢٤,٨٦%) إيجابية الأجسام المضادة (IgG) . هذه هي الدراسة الأولى من نوعها وتؤكد صلاحية طريقة الميا لإجراء دراسات وبائية من هذا النوع.