Journal of the Egyptian Society of Parasitology, Vol. 47, No. 3, December 2017
J. Egypt. Soc. Parasitol. (JESP), 47(3), 2017: 689– 694

SERO-PREVALENCE OF TOXOPLASMOSIS AMONG MULTIPAROUS PREGNANT WOMEN ATTENDING ANTENATAL CARE AT BENI-SUEF UNIVERSITY'S HOSPITAL, EGYPT

BY

SAMAH S. ABDEL GAWAD[1], MARWA A. GHIETH[1]*, NESREEN M. KAMEL[2] AND SHERWET M. SHAWKEY[3]

Department of Medical Parasitology[1], Department of Clinical Pathology[2], and Department of Obstetrics and Gynecology[3], Faculty of Medicine, Beni-Suef University, Beni-Suef 62511, Egypt (*Correspondence: marwaghieth@yahoo.com)

Abstract

Toxoplasma gondii is a critical zoonotic protozoan infection associated with comorbidity on pregnancy and fetus. This study evaluated over a year the sero-prevalence of *T. gondii* among multiparous pregnant women undergoing antenatal care at Beni-Suef University's Hospital, and estimated the risks for infection. All patients were subjected to immunological detection of both IgG & IgM anti-toxoplasmosis antibodies using ELISA. Out of 300 females, anti-*T.gondii* antibodies were detected in 20% (n=60) of cases. Of 60 seropositive cases, IgG+ve, IgM+ve and both IgG+ve and IgM+ve were detected in 56.6% (n=34), 23.3% (n=14) and 20% (n=12) respectively. Positive cases were common in age groups 30-40 (45%) and 20-30 (43.3%). 50% of positive patients suffered from first trimester abortion (p=0.005). History of the previous obstetric outcome was a predictive factor (p =0.002) for infection. None of other variables were risks.

Key words: Egypt, Toxoplasmosis *gondii*, Seroprevalence, Pregnancy, ELISA,

Introduction

Toxoplasma gondii is an obligate intracellular protozoon that infects warm blooded animals including human. About 30% of people are chronically infected worldwide (Liu *et al*, 2009). While human infection can occur via ingestion of tissue cysts from infected meat, most human infection comes from oocysts, cats are the only definitive host, and thus shedding of oocysts by cats provides the ultimate source of toxoplasmosis (Elsheikha and Morsy, 2009). So, felines serve as definitive hosts, while all other vertebrates, including man act as intermediate hosts of the parasite with disseminated tissue infections (Lilly and Wortham, 2013). The parasite is distributed worldwide and has significant impact not only on animal production but also on public health worldwide (Bamba *et al.*, 2017). *T. gondii* can cross the placenta causing congenital toxoplasmosis with a worldwide distribution (Eissa *et al*, 1990; Bamba *et al*, 2017).

The infection may lead to spontaneous abortion, stillbirth, neonatal death, premature labor or congenital anomalies (Sudan *et al*, 2013), as well as serious progressive visual, hearing, motor, cognitive and neurological child problems (Peyron *et al*, 2015). Thus, rapid diagnosis of acute infection during pregnancy proved essential to assess risk of vertical transmission and prevention of expected complications (Saki *et al*, 2015) using immuno-assay for detection of toxoplasmosis antibodies proved to be simple and helpful (Kim *et al*, 2017).

The present study aimed to detect the seroprevalence of *T. gondii* among out-patient pregnant women attending the antenatal care unit, Beni-Suef University's Hospital.

Subjects, Materials and Methods

Study design and population: A cross sectional study was performed over a year on 300 pregnant women attending Beni-Suef University Hospital for antenatal care. Target population was multiparous pregnant women with history of abortion or full term baby. Women with history of clinical systemic or physical causes of fetal wastage, hypertension, diabetes or Rh incompatibility HIV patients or infected with any autoimmune disease that interferes with antibodies detection were excluded. Patients were divided according to previous pregnancies outcome into four groups.

GI: included women with history of 1st trimester abortion (≤12 weeks gestation), GII: women with history of 2nd trimester abortion (13-26 weeks gestation), GIII: women with history of intrauterine fetal death and GIV, women with history of normal full term baby as a control.

Ethical guidelines: Patients were informed about the study purpose and its consequences. Samples collection wasn't done except by their approval which was in accordance with the 1964 Helsinki declaration.

Data collection: Predesigned questionnaire sheets were filled out on each patient including sociodemographic data as age, residence, education level, occupation and obstetric history.

Samples: Venous blood samples were collected, left to stand, centrifugation at 3000 rpm for 4 minutes and then serum was separated and stored in a labeled clean Eppendorf tubes at -20°C till needed.

Immunoassay: Sera were measured for ELISA IgG & IgM using inactivated antigen by using NovaLisa™ ELISA kits (Nova Tec Immundiagnostica GmbH, Dietzenbach/Germany) after the manufacturers' instructions.

Statistical analysis: Data were analyzed using SPSS software version 18, (Chicago, Illinois, USA). Frequency distribution and descriptive statistics were presented by M±SD. Univariate analysis detected risk factors associated with positive anti-*T. gondi* antibodies. Comparison was done using Chisquare and t-test. *P* less than 0.05 were significant.

Among 300 antenatal care multiparous women with age ranged from 15 to 40 years old (27.9±7.1), *T. gondii* antibodies were detected in 60 (20%) cases. They were Ig G+ve (56.6%), Ig M+ve (23.3%) and both Ig G+ve and Ig M+ve (20 %). Old toxoplasmosis (only IgG+ve) was highest in GIII and GIV (80% & 58.8%, respectively) & low in GII (50%). Acute infection (only IgM+ve) was high in GI & G IV (26.6% & 23.5%, respectively). Acute infected women (IgG+ve & IgM+ve) were common in GII (37.5%) and GI (20%).

Characteristics and risk factors for seropositive cases: As to age groups, early and late young pregnant women (20-30 years) and (30-40 years) represented the high seropositive groups (43.3% & 45% respectively), pregnant women with 40 years was the lowest (5%). Most of toxoplasmosis seropositive patients were at the 3rd and 2nd trimester during the study (56.7% & 41.7%, respectively) and only one patient was at the 1st trimester (1.6%). Exactly 50% of seropositive patients were complaining of history of abortion during the first trimester and only 8.3% with history of intrauterine foetal death. Positive cases were from rural area (63.3%) and with different jobs and different education level. None of these variables proved risk factor, only history of obstetric outcome was a significant risk (p =0.002), and spontaneous abortion during 1st trimester was associated with positive cases (p = 0.005). The details were given in tables (1, 2 & 3).

Results

Table 1: Socio-demographic, clinical and immunological variables among population

Characteristics	Patients (n= 300) (%)
Age (M±SD) years	27.9±7.1
Gestation age of population 1st trimester	2 (0.7)
2nd trimester	140 (46.7)
3rd trimester	158 (52.7)
History of obstetric outcome 1.Full term baby	104 (34.7)
2.First trimester abortion	31 (10.3)
3.Second trimester abortion	15 (5)
4.Intra uterine foetal death	150 (50)
Seropositive for toxoplasmosis antibodies	60 (20)

Table 2: Immunological prevalence of *toxoplasmosis* among antenatal care women.

Groups	Seropositive for toxoplasmosis antibodies (n=60)				*P* value
	IgG+ve (n) (%)	IgM +ve (n) (%)	IgG+ve & IgM+ve (n) (%)	Total	
First trimester abortion (n=104)	16 (53.3)	8 (26.6)	6 (20)	30 (50)	0.005[a]
Second trimester abortion (n=31)	4 (50)	1(12.5)	3 (37.5)	8 (13.3)	0.300
Intra uterine foetal death (n=15)	4 (80)	1(20)	0 (0)	5 (8.3)	0.108
Full term baby (n=150)	10 (58.8)	4 (23.5)	3 (17.6)	17 (28.3)	0.100
Total	34 (56.6)	14 (23.3)	12 (20)	60 (100)	

[a]Significant $P < 0.05$

Table 3: Socio-demographic and clinical variables distribution among toxoplasmosis seropositive women

Variables	Toxoplasmosis seropositive (n=60) (%)	Toxoplasmosis sernegative (n=240) (%)	*P* value
Age (M± SD) years	29.25±7.15	27.39±7.16	0.951
Age groups			
1.Teen age (>15-20)	4 (6.7)	46 (19.2)	0.08
2.Early Young (>20-30)	26 (43.3)	102 (42.5)	
3.Late young (>30-40)	27 (45)	86 (35.8)	
4.Early Adulthood (=40)	3 (5)	6 (2.5)	
Gestation age			
1st trimester	1 (1.6)	1 (0.4)	0.416
2nd trimester	25 (41.7)	115 (47.9)	
3rd trimester	34 (56.7)	124 (51.7)	
History of obstetric outcome			
1.Full term baby	17 (28.3)	133 (55.4)	0.002[a]
2.First trimester abortion	30 (50)	74 (30.8)	
3.Second trimester abortion	8 (13.3)	23 (9.6)	
4.Intra uterine foetal death	5 (8.3)	10 (4.2)	
Residence			
Rural	38 (63.3%)	161 (67.1%)	
Urban	22 (36.7%)	79 (32.9%)	0.343
Occupation			
Workers	24 (40.0%)	86 (35.8%)	
Housewives	22 (28.2)	92 (24.4)	0.736
Employees	10 (16.7)	54 (22.5)	
Farmers	4 (6.7)	8 (3.3%)	
Education level			
Illiteracy	10 (16.7)	30 (12.5)	0.083
Basic and secondary	22 (36.7)	59 (24.6)	
High school	17 (28.3)	55 (22.9)	
University	11 (18.3)	96 (40)	
Season			
Spring (March-May)	10 (16.7)	53 (22.1)	0.517
Summer (Jun-August)	24 (40)	107 (44.6)	
Autumn (September-November)	15 (25)	45 (18.8)	
Winter (December-February)	11 (18.3)	35 (14.6)	

[a]Significant *P*-value < 0.05

Discussion

In the present study, ELISA-antibodies against *T. gondii* among antenatal care women were 20%. Higher prevalence rates (64.7%) were reported at El Menoufia (El Deeb *et al*, 2012) and El Minia (50.8%) Governorates (Kamal *et al*, 2015), but Saleh *et al.* (2014) at Alexandria reported 22.2% and 20% prevalence rates by antibody detection against *T. gondii* among pregnant and non-pregnant women.

Generally, difference in seroepidemiology of toxoplasmosis between studies might be attributed to sensitivity of the different used tests, susceptibility of the studied populations, environmental conditions and domestic animals number, different culture of each community with meat consumption and food processing. Some authors (Abd El-Razik *et al*, 2014) reported that, consumption of outdoor improbably cooked meat by Egyptian families at junk food shops as a main cause

of high serological prevalence of toxoplasmosis.

In the present study, women with IgG+ve represented 56.6% of cases, IgM+ve represented 23.3%, and 20% were IgG & IgM positive. Hussein *et al*. (2001) in Qualyobia reported seropositivity to anti-*Toxoplasma* IgG in random (57.9%), full term (58.1%) and aborted samples (44.7%). Tammam *et al*, (2013) in Qena reported seropositivity rates of 46.1%, and 18.4% for IgG & IgM, respectively, among women with spontaneous abortion.

In the present study, immunized women with IgG+ve had no impact risk on their fetus. However, latent toxoplasmosis could not be excluded as a cause of their previous abortion. Since the outcome of toxoplasmosis depended on the immunity state of the mother and time of acquiring primary infection (Tenter *et al*, 2000). Among immunocompetent mother, it's at least 4-6 months or more from getting primary infection before conception to reach a satisfied level of protective immunity to the baby (Ebbesen *et al*, 2000).

In the present study, positivity to IgM was 23.3% of cases, these women were more susceptible to have bad impact on their fetus as they acquires primary infection during pregnancy and they were IgG–ve. Treatment of toxoplasmosis in these women lowed risk of congenital toxoplasmosis (McLeod *et al*, 2000). However, the authors couldn't depend on IgM anti-*Toxoplasma* antibody alone to diagnose active infection as it can stay elevated for months after infection, raising IgG titer was needed, in addition to the fact that, commercial IgM diagnostic kits could yield a number of many false-positive results (Dhakal *et al*., 2015).

Spontaneous abortion during the 1st trimester was significantly associated with positive cases (p =0.005) and IgM+ve was 26.6% among them. This result agreed with El-Gazamy *et al*, (2009) and Tammam *et al*, (2013) Acquiring primary infection during the 1st trimester carried high risk on the outcome (Sahwi *et al*, 1995; Gilbert *et al*, 2000). Some authors (Wang *et al*, 2003; Wyatt *et al*, 2005) ensured that, spontaneous abortion occurs in the first trimester and frequently decreases with progress of pregnancy. Attributing this to hormonal factor, estrogen and progesterone deviation later in pregnancy (El-fadaly *et al*., 2012).

In the present study seropositivity showed two peaks during summer (40%) and autumn (25%), without risk factor for infection (p >0.005). In tropics area the epidemiology of toxoplasmosis was linked to seasons. Climate conditions proved to be vital for oocyst sporulation, worm and humid weather during summer and early autumn help in survival and accumulation of oocyst than cold areas (Meerburg and Kijlstra, 2009). Weather changes could alter cycles of infection transmission through pathogen survival and water born dynamics for transmission (Patz *et al*, 2008; Plowright *et al*. 2008).

To lower congenital infection, pregnant women should be informed about the risks of acquiring toxoplasmosis during pregnancy (Liu *et al*., 2009). In addition to risk factors; consumption of raw vegetables, undercooked meat and kitten's exposure are crucial risks for *T. gondii* infection (Jones *et al*. 2009). As millions of oocysts are shed by cats after getting infected and survive for long time (Dabritz *et al*., 2007). The role of mice in disease cycle couldn't be ignored, as a cat source of infection through preying on infected mice, thus prevention of infection includes infected mice control (Jiang *et al.*, 2012). *T. gondii* find their way in goat's milk and fresh cheese as mentioned by Dubey *et al*. (2014).

In the present study seropositive cases to *T. gndii* antibodies were common among late and early young women (45% and 43.3%, respectively). Many studies (Sarkar *et al*, 2012; Chintapally *et al*, 2013; Kamal *et al*, 2015) observed seropositivity in the age group of 21-30 years. Infection was common among rural residence (63.3%), workers (40.0%), and women attending

basic and secondary schools (36.7%). But, none were risks for infection.

Seronegative mothers (80%) detected in the present study were at risk of seroconversion by acquiring the infection during pregnancy. Thus, they should undergo follow up for anti-toxoplasmosis antibodies each trimester and education program to avoid infection.

Conclusion

Infection with *Toxoplasma gondii* is common, but it can have serious consequences in the pregnant women if passed to the developing fetus.

To carry out anti-*Toxoplasma* antibodies as a screening test for all females before and after conception is essential at Beni-Suef City. Follow up proved essential for pregnant women by doing simple antibody detection test to avoid sero-conversion. Educational health programs are needed among females informing them about toxoplasmosis as a risky congenital transmitted disease and preventive measures.

Acknowledgments

The authors are would like to thanks the Scientific Research Developing Unit, Beni-Suef University for allowing and facilitating this study. The fund was kindly given to the first author, Dr. Samah S Abdel Gawad. Thanks are also extended to the patients who accepted to share in the practical study

Conflict of interest: The authors declared that had no conflict of interest except scientific the aspect.

References

Abd El-Razik, KA, El Fadaly, HA, Barakat, AMA, Abu Elnaga, ASM, 2014: Zoonotic hazards *T. gondii* viable cysts in ready to eat Egyptian meat-meals. World J. Med. Sci. 11, 4:510-7.

Bamba, S, Cissé, M, Sangaré, I, Zida, A, Ouattara, S, *et al*, 2017: Seroprevalence and risk factors of *Toxoplasma gondii* infection in pregnant women from Bobo-Dioulasso, Burkina Faso. BMC Infectious Diseases 17:482. doi:10. 1186/s12879-017-2583-6.

Chintapally, S, Padmaja, IJ, 2013: Seroprevalence of toxoplasmosis in antenatal women with bad obstetric history. Trop. Parasitol. 3:62-6.

Dabritz, HA, Miller, MA, Atwill, ER, Gardner, IA, Leutenegger, CM, *et al*, 2007: Detection of *Toxoplasma gondii*-like oocysts in cat feces and estimates of the environmental oocyst burden. J. Amer. Vet. Med. Assoc. 23:1676-84.

Dhakal, R, Gajurel, K, Pomares, C, Talucod, J, Press, CJ, *et al*, 2015: Significance of a positive *Toxoplasma* immunoglobulin M test results in the United States. J. Clin. Microbiol. 53:3601-5.

Dubey, JP, Verma, SK, Ferreira, LR, Oliveira, S, Cassinelli, AB, *et al*, 2014: Detection and survival of *Toxoplasma gondii* in milk and cheese from experimentally infected goats. J. Food Prot. 77, 10:1747-53

Ebbesen, P, 2000: Placenta physiology. In: Ambroise-Thomas, P, Petersen, E, Congenital Toxoplasmosis: Scientific Background, Clinical Management and Control. Paris: Springer-Verlag.

Eissa, MH, Abdel Salam, AM, Herez, HA, Younis, TA, Morsy, TA, 1990: Placental villous maturation in patients with repeated abortions and chronic toxoplasmosis. J. Egypt. Soc. Parasitol. 20, 2:661-6.

El Deeb, HK, Salah-Eldin, H, Khodeer, S, 2012: Prevalence of *Toxoplasma gondii* infection in antenatal population in Menoufia Governorate, Egypt. Acta. Trop.124: 185-91.

El-Fadaly, HA, Hassanain, MA, Shaapan, R M, Barakat, AM, Toaleb, NI, 2012: Serological and hormonal assays of murine materno-fetal *Toxoplasma gondii* infection with emphasis on virulent strains. World J. Med. Sci. 7, 4:248-54.

El-Gozamy, BR, Mohamed, SA, Mansour, H A, 2009: Toxoplasmosis among pregnant women in Qualyobia Governorate, Egypt. J. Egypt Soc. Parasitol. 39, 2:389–401.

Elsheikha, HM, Morsy, TA, 2009: Role of immune response in *Toxoplasma gondii* tachyzoite-bradyzoite stage interconversion: A Janus in determining disease outcome. J. Egypt. Soc. Parasitol. 39, 2:595-8.

Gilbert, R, 2000: Epidemiology of infection in pregnant women. In: Congenital Toxoplasmosis: Scientific Background, Clinical Management and Control. Ambroise-Thomas, P, Petersen, E, (eds.). Paris: Springer-Verlag.

Hussein, AH, Ali, AE, Saleh, MH, Nagaty, I M, Rezk, AY, 2001: Seroprevalence of *Toxoplasma* infection in Qualyobia Governorate, Egypt. J. Egypt Soc. Parasitol. 31:355-63.

Jiang, W, Sullivan, AM, Su, C, Zhao, X, 2012: An agent-based model for the transmission dyn-

amics of *Toxoplasma gondii*. J. Theor. Biol. 293: 15-26.

Jones, JL, Dargelas, V, Roberts, J, Press, C, Remington, JS, *et al*, 2009: Risk factors for *Toxoplasma gondii* infection in the United States. Clin. Infect. Dis. 49, 6:878-84.

Kamal, AM, Ahmed, AK, Abdellatif, MZM, Tawfik, M, Hassan, EE, 2015: Seropositivity of toxoplasmosis in pregnant women by ELISA at Minia University Hospital, Egypt. Korean J. Parasitol. 53, 5:605-10.

Kim, YH, Lee, J, Hoo-Ahn, S, 2017: Seroprevalence of toxoplasmosis with ELISA and rapid diagnostic test among residents in Gyodong-do, Inchon City, Korea: A four-year follow-up. Korean J. Parasitol. 55, 3:247-254.

Lilly, EL, Wortham, CD, 2013: High prevalence of *Toxoplasma gondii* oocyst shedding in stray and pet cats (*Felis catus*) in Virginia, United States. Parasit. Vectors Sept. 17. 6:266. doi: 10 1186/1756-3305-6-266.

Liu, Q, Wei, F, Gao, S, Jiang, L, Yuan, B, *et al*, 2009: *Toxoplasma gondii* infection in pregnant women in China. Trans. R. Soc. Trop. Med. Hyg. 103:162-6.

McLeod, R, Remington, JS, Behrman, RE, Kliegman, RM, Jenson, HB, 2000: Toxoplasmosis (*Toxoplasma gondii*): Nelson Textbook of Pediatrics. 16th edition. Philadelphia, Pennsylvania, WB Saunders USA.

Meerburg, BG, Kijlstra, A, 2009: Changing climate changing pathogens: *Toxoplasma gondii* in North-Western Europe. Parasitol. Res. 105: 17-24

Patz, JA, Olson, SH, Uejio, CK, Gibbs, HK, 2008: Disease emergence from global climate and land use change. Med. Clin. North America 92:1473-91.

Peyron, F, Wallo, NM, Kieffer, F, Garweg, J G, 2015: Toxoplasmosis. In: Remington and Klein's infectious diseases of the fetus and newborn infant Wilson, CB, Nizet, V, Maldonado, Y, Remington, JS, Klein, JO (eds.). Elsevier Health Sciences, Oxford, United Kingdom.

Plowright, RK, Sokolow, SH, Gorman, ME, Daszak, P, Foley, JE, 2008: Causal inference in disease ecology: investigating ecological drivers of disease emergence. Frontiers Ecol. Environ. 6:420-9.

Sahwi, SY, Zaki, MS, Haiba, NY, Elsaid, OK, Anwar, MY, Abd-Rabbo, SA, 1995: Toxoplasmosis as a cause of repeated abortion. J. Obstet. Gynaecol. 21, 20:145-48.

Saki, J, Mohammadpour, N, Moramezi, F, Khademvatan, S, 2015: Seroprevalence of *Toxoplasma gondii* in women who have aborted in comparison with the women with normal delivery in Ahvaz, Southwest of Iran. Scientific World J., ID 764369, 4. doi:10.1155/2015/764369.

Saleh, AMA, Ali, HA, Ahmed, SAM, Hosny, S M, Morsy, TA, 2014: Screening of *Toxoplasma gondii* infection among childbearing age females and assessment of nurses' role in prevention and control of toxoplasmosis. J. Egypt. Soc. Parasitol. 44, 2:329-34.

Sarkar, MD, Anuradha, B, Sharma, N, Roy, RN, 2012: Seropositivity of toxoplasmosis in antenatal women with bad obstetric history in a tertiary-care hospital of Andhra Pradesh, India. J. Health Popul. Nutr. 30: 87-92.

Sudan, V, Jaiswal AK, Shanker, D, 2013: Recent trends in the diagnosis of toxoplasmosis. Clin. Rev. Opinions 5, 2:11-7.

Tammam, AE, Haridy, MA, Abdellah, AH, Ahmed, SR, Fayed, HM, Alsammani, MA, 2013: Seroepidemiology of *Toxoplasma gondii* infection in women with first trimester spontaneous miscarriage in Qena Governorate, Egypt. J. Clin. Diag. Res. 7, 12:2870-3.

Tenter, AM, Heckeroth, AR, Weiss, LM, 2000: *Toxoplasma gondii*: from animals to humans. Int. J. Parasitol. 30:1217-38.

Wang, X, Chen, C, Wang, L, Chen, D, Guang, W, *et al*, 2003: Conception, early pregnancy loss and time to clinical pregnancy: A population-based prospective study. Fertil. Steril. 79:577-84.

Wyatt, PR, Owolabi, T, Meier, C, Huang, T, 2005: Age-specific risk of fetal loss observed in the second trimester serum screening population. Am J. Obstet. Gynecol. 192:240-6.