

Prevalence of IgG antibodies against *Toxoplasma gondii* among Syrian females of childbearing age

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Toxoplasma gondii is an intracellular parasite that can infect almost all mammals and various species of birds. It is also considered one of the most widespread parasite agents among humans with an estimated one third of population possibly parasitized. This parasite is the causative agent of toxoplasmosis which healthy humans, is generally sub-clinical, or mild. However, this infection can cause significant morbidity and mortality in the developing fetus if the mother acquires the primary infection during pregnancy. With increasing gestational age, transmission rate to the fetus increases while the severity of congenital disease decreases. The spectrum of outcome of congenital toxoplasmosis ranges from death in utero, chorioretinitis or hydrocephalus with mental impairment, to a healthy infant without clinical signs of toxoplasmosis. The symptoms may be present at birth, or develop later in life, leading to retinal diseases and blindness, psychomotor retardation, neurological abnormalities or hearing difficulties. Prevention of human toxoplasmosis relies on the knowledge of the disease epidemiology.¹ However, the life cycle of *Toxoplasma gondii* is complex since virtually all mammals and birds may serve as intermediate, or definitive hosts, with cats being the predominant definitive host. Post-natal human infection may result from ingestion of tissue cysts contained in raw, or undercooked meat from intermediate hosts or from ingestion or inhalation of oocysts shed by definitive hosts and disseminated in soil and water contaminating fruits and vegetables. Therefore, incidence and prevalence vary among countries, and even among regions within countries. Since only infection during pregnancy can cause threat to humans, studies are mostly interested on the determination of epidemiological features of *Toxoplasma* infection among women either before or during pregnancy. Detection of toxoplasmosis is currently dependent on serological techniques screening for IgM and IgG antibodies, the former indicating recent infection while the latter indicating a past exposure and the existence of a protective immunity. In Syria, there is a lack of data available concerning the seroprevalence of *Toxoplasma* antibodies. With the lack of such studies, the magnitude of toxoplasmosis on fetuses is still unknown. Therefore, the objective of the study was to determine the current *Toxoplasma* IgG antibodies circulation among Syrian females at childbearing age, and to investigate factors associated with seropositivity.

A cross-sectional design was used to evaluate seroepidemiology of *Toxoplasma* IgG among Syrian females at childbearing age. The study participants were female students attending the Pharmacy College in the University of Kalamoon, Deratiah, Syria. Eligible participants were those aged between 18-40 years old, and not previously pregnant. Students who consented to participate in the study were asked to give a blood sample and provide information on a) demographic data, such as name, age and marital status; b) exposure to contamination by food, such as eating undercooked, or uncooked meat, eating fruits and vegetables without good washing (assessed as yes or no); c) contact with pet animals, such as having or caring for a cat (assessed as yes or no); and d) exposure to soil, such as having a garden, or agriculture hobby (assessed as yes or no).

The project was approved by the Pharmacy College Ethical Committee, University of Kalamoon, Deratiah, Syria. The sera were collected between March 2008 and May 2008 and tests were conducted in the Microbiology Laboratory in the Faculty of Pharmacy, University of Kalamoon. Samples were tested for *Toxoplasma* specific IgG antibodies using an enzyme linked immunoassay for *Toxoplasma* IgG detection by Diamed Eurogen, 2300 Turnhout, Belgium. *Toxoplasma* IgG titers greater than 25 IU/ml were considered positive; those between 20-25 IU/ml were equivocal, and those less than 20 IU/ml were considered negative. All samples were tested twice. Data were analyzed using the Statistical Package for Social Sciences (SPSS Inc., Chicago, IL, USA) Version 13. The Fisher Exact Test was employed to analyze the difference between positive samples and negative samples on factors associated with seropositivity. Statistical significance was set at $p < 0.05$.

At the time of samples collection (March - May 2008), there were 376 registered female students in the Faculty of Pharmacy in the University of Kalamoon. A total of 104 students agreed to participate in the study, with a response rate of 27.65%. Of the 104 samples collected, 90 were tested for *Toxoplasma* specific IgG using the ELISA method. The other 14 samples were excluded from analysis due to hemolysis of the blood and insufficient data from participants. The majority of participants were 22 years old, or below (88%), single and living in various areas of Syria.

Analysis revealed that 26.7% (24 samples) of the cohort was seropositive (positive sample); while a relatively high proportion of participants (73.3%; 66 samples) were *Toxoplasma* susceptible subjects (negative sample). The variability in the positive values of antibodies ranged between 44.8-750 IU/ml. There were no participants with antibodies values close to the cut-off of 25 IU/ml. A clear majority of the positive

Table 1 - Number of participants exposed to different sources of contamination in the positive and negative sample, n(%).

Sources	Total sample (n=90)	
	Positive (n=24)	Negative (n=66)
Having, or caring for pet animals (cats)*	11 (46)	12 (18)
Eating uncooked or undercooked meat	15 (63)	36 (55)
Eating fruits and vegetables without good washing	10 (42)	21 (32)
Gardening or agricultural hobbies*	9 (38)	10 (15)

n - number of participants, *indicates statistical significance

sample registered values of antibodies above 120 IU/ml; while only 2 participants had antibodies values below 100 IU/ml.

Table 1 shows the number of participants exposed to different sources of infection of *Toxoplasma* (eating raw, or undercooked meat, gardening, or having agricultural hobbies and keeping pet animals), which were considered factors that could be associated with seropositivity. Comparisons of the positive sample with the negative sample groups were made on these factors. Results showed that there were significant differences among the 2 groups (positive sample and negative sample) with respect to having, or caring for a pet animal (cat) ($p=0.013$) and gardening, or agricultural hobbies ($p=0.038$). Significantly, more students were caring for a pet animal (cat) and were gardening, or had agriculture as a hobby in the positive sample group compared with those in the negative sample group. There was a trend, although this did not reach statistical significance, towards more students consuming raw, or undercooked meat and eating vegetables and fruits without good washing in the positive sample group compared with the negative sample group.

Our study investigates the seroprevalence of *Toxoplasma* IgG antibodies among females of childbearing age in Syria. We approached female students attending a Syrian University in order to screen for *Toxoplasma*-specific IgG. Our findings showed that only 26.7% of the subjects tested were found to have IgG against the *Toxoplasma* parasite. This immunity may have been acquired from previous infection with the parasite during childhood, or adolescence. Among the participants, a high percentage (73.3%) did not have sufficient immunity against *Toxoplasma gondii*. Our results agree with the regional and universal trend for toxoplasmosis infection ratio, where a quarter to one third of various populations showed immunity.^{2,3}

Our results demonstrated a significant difference in seropositivity in relation to contact with pets (cats) and gardening, or agricultural hobbies. Although this failed to reach statistical significance, there was a trend towards more students in the positive sample group eating raw,

or undercooked meat and eating fruits and vegetables without adequate washing. Considering the high spread of domestic and stray cats in Syria, the consumption of uncooked vegetables and the suitable climatic conditions for sporulation of *Toxoplasma gondii* oocysts, the exposure to cat feces seems to be the principal route for *Toxoplasma* infection in most parts of the country. Consumption of undercooked meat (Kibbeh nayyeh) could also be suggested as a second possible means for acquiring *Toxoplasma* infection in Syria.⁴

The major limitation of our study was the low number of samples tested, which precluded the use of sophisticated statistical analysis. Also, we tested a specific population of participants, such as single individuals, females, and university students. Therefore, our findings may not generalize to all the female Syrian population. However, since there is an evident lack of research concerning this issue, our study constitutes an essential preliminary investigation in Syria, which draws attention to the fact that the risk of acquiring the infection for the first time among females at childbearing age is still high.

Systematic screening programs such as TORCH test in countries such as France, where serological surveillance of seronegative pregnant women for seroconversion is compulsory, have indicated that 1-1.5/1,000 newborns suffer from congenital toxoplasmosis, while in the USA, the incidence of acute *Toxoplasma* infection during pregnancy has been estimated at 0.2-1%, and the incidence of congenital infection ranging from 1/1000 to 1/8000 live births. In some European countries, where educational measures have been incorporated into routine obstetric care, reductions in incidence of infection by nearly 50% have been reported.⁵ Therefore, screening of pregnant women for the *Toxoplasma* antibody and educating them on the sources of transmission and preventive measures for disease should be essential protocols, that we would suggest for a country with a high prevalence of seronegativity such as Syria. In our study, women who were negative for the antibodies were educated regarding toxoplasmosis, and on the importance of testing, especially during a future pregnancy.

In conclusion, our study shows a relatively high percentage of unprotected women of childbearing age against *Toxoplasma*. We strongly recommend that larger studies are performed in the future, targeting different demographic groups, namely, a broader age range, and factors associated with seropositivity. This would allow a more detailed statistical analysis, using multivariate analysis, which could provide important insights regarding the *Toxoplasma* antibodies circulation among Syrian females of childbearing age. Also, findings from

these studies would allow the design of interventions to be targeted at high risk individuals. We recommend that screening women prior or during pregnancy is important to describe the immune status towards Toxoplasma and hence determine the outcomes if any new infection occurs during pregnancy.

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