



Sero-epidemiology of human *Toxoplasma gondii* infection among male population in Charsadda, KPK, Pakistan

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Abstract

Toxoplasmosis is a zoonotic infection of humans and animals which is caused by the opportunistic protozoan *Toxoplasma gondii*. The parasite belongs to the phylum Apicomplexa. Worldwide one-third of the total population are chronically infected by the parasite. Clinically the parasitic infection produces a wide range of symptoms in the humans. The deteriorating health impact includes blindness and mental retardation in congenitally infected children. The aim of the current study was to determine the seroprevalence of *T. gondii* among male population in District Charsadda, KPK. 300 random samples were collected from the male population. Serum was extracted from the samples and was subjected to the serological technique. The detection was done with Latex Agglutination kit. Out of 300 samples, 63 (21%) were found positive for the parasite. A significant association was determined between the infection and age group in the population. From results, it was concluded that the infection was more prevalent in the age group 46-55 years with 35.41% followed by the age groups 66-75 years, 56-65 years, 36-45 years, 26-35 years and 15-25 years with 33.33%, 30.55%, 22.95%, 18.46% and 8.33% respectively. Bringing awareness to the public and educate them about the health-associated risks upsurging from unhealthy food and lifestyle habits. In the future updates of the global burden of infections, the data on Toxoplasmosis epidemiology and its transmission should be included in order to support public health interferences to eradicate the disease burden.

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Introduction

Toxoplasmosis is a zoonotic infection which is caused by an intracellular protozoan parasite *Toxoplasma gondii*. The parasite belongs to the phylum Apicomplexa. Other members of the phylum include Plasmodium, Neospora, and Sarcocystis. The phylum is very vast having diverse groups of parasites capable of causing life-threatening infections in human and animals (Majumdar 2010). Variation in the *Toxoplasma* species is dependent on several factors such as age, nutritional and sociocultural habits, climate, geography, and contact with domestic animals (Al-Saadii 2013). *T. gondii* infection is highly prevalent and it was estimated that the infection frequency ranges from 5 to 95% in various regions of the world. It was recognized as one of the most prevalent human parasitic infection in the world. Clinical diagnosis of the early infection is still a very concerning issue (Chintapalli and Padmaja 2013).

Epidemiological reports show that worldwide *T. gondii* affects approximately 30-50% of the human population. The infection is common in both developed and developing nations (Flegr, Prandota *et al.* 2014). *T. gondii* is a parasite having the capability of penetration into several host cells (LEVINE 1977). Surveys based on the serological examination of the infection have been conducted in various parts of the world and has reported the occurrence of anti-*T. gondii* antibodies in more than one-third of the human population (Ashburn 1992). The infection poses a serious concern in case of immunosuppressed patients i.e. patients having to undergo immunosuppressive cancer therapy and also during organ transplantation (Derouin and Pelloux 2008).

Worldwide one-third of the population is affected by the parasitic infection. However, the frequency of the affected population is not same all over. The risk for occurrence of the infection is directly associated with time because it is a lifelong infection. Therefore the prevalence of the parasitic infection is majorly dependent on the age variable of patients (Tenter, Heckeroth *et al.* 2000). First epidemiological study on the prevalence of *T. gondii* infection was reported in 1951 by Zeipel and Linder. In that study 300 healthy blood donors were screened for the parasitic infection

and prevalence was estimated to be around 37% (Zeipel and Linder 1951). Another study conducted by Hedqvist in 1953 showed the frequency of the infection in various age groups in Eskilstuna.

The infection was found to be most prevalent in the age group 20-50 years with frequency of 40% as compared to 0% prevalence in the age group 0-4 years (Hedqvist 1953). The aim of the current study was to determine the seroprevalence of Toxoplasmosis infection in male population in Charsadda, KPK Pakistan.

Materials and methods

Subjects

The current study was conducted in district Charsadda, KPK. The samples were collected from 3 tehsils of the district including Charsadda, Tangi, and Shabqadar. A total of 300 random samples were collected from the male population. To determine the seroprevalence of *T. gondii*, 100 samples were collected from each Tehsil. The sampling was done from May 2017 to July 2017. The samples were tested for the parasite at Mardan Medical Complex. The age of the participants varied from 15-70 years.

Serological technique

The detection of the *T. gondii* parasite was done with "Latex Agglutination Kit". Blood was taken from the participants and was centrifuged at 8000rpm for 1min. The serum was transferred to fresh microfuge tubes. 50µl of the serum was added to fresh tube followed by the addition of 25µl of latex reagent. The tubes were left at room temperature for 3min. The agglutination of the serum samples indicated the presence of the parasite while smooth suspension shows a negative result.

Statistical analysis

Data analysis was done by Epi Info 7 (Center for Disease Control, GA, USA). The data was analyzed on the basis of tehsil, age group, location, residence type, marital status, hygienic condition and routine contact with domestic animals.

Results

Overall Seroprevalence of *T. gondii*

Out of 300 samples, 63 (21%) were found positive for *T. gondii* parasite. The remaining 237 samples were

negative because no agglutination occurred in the serum.

The results are shown in Table 1.

Table 1. Seroprevalence of *T. gondii* in male population.

Total samples (n)	Positive	Negative	Prevalence %
300	63	237	21%

Seroprevalence of *T. gondii* in different Tehsils

The prevalence of *T. gondii* parasite was determined from different Tehsils included in the study. As 100 samples were collected from each Tehsil in which the

prevalence of the infection was as follows; 17 (17%) were found positive from Tehsil Charsadda, 23 (23%) positive from Tehsil Tanggi and 23 (23%) found positive from Tehsil Shabqadar as shown in Table 2.

Table 2. Tehsil wise seroprevalence of *T. gondii*.

Tehsil name	Total samples	Positive	Negative	Prevalence %
Tehsil Charsadda	100	17	83	17%
Tehsil Tanggi	100	23	77	23%
Tehsil Shabqadar	100	23	77	23%

Seroprevalence of *T. gondii* in different age groups

The age of the participants varied from 15 to 75 years. The studied population was divided into different age groups and in each group, the frequency of the infection was determined. 7 (8.33%) out of 84 were found positive for the parasite which belongs to the age group 15-25 years. 12 (18.46%) out of 65, 14 (22.95%) out of 61, 17 (35.41%) out of 48, 11 (30.55%)

out of 36 and 2 (33.33%) out of 6 were found positive in the age groups 26-35, 36-45, 46-55, 56-65 and 66-75 years respectively. The infection was highly prevalent in the age group 46-55 years with 35.41% followed by age groups 66-75 years with 33.33%, 56-65 years with 30.55%, 36-45 years with 22.95%, 26-35 years with 18.46% and 15-25 years with 8.33% as shown in Table 3.

Table 3. Age wise seroprevalence of *T. gondii*.

Age groups (Years)	Total samples (n)	Positive	Prevalence %
15-25	84	7	8.33%
26-35	65	12	18.46%
36-45	61	14	22.95%
46-55	48	17	35.41%
56-65	36	11	30.55%
66-75	6	2	33.33%

Seroprevalence of *T. gondii* on the basis of the hygienic condition

The seroprevalence of the parasite was determined on the basis of the hygienic condition of the studied population. The participants were from different hygienic environment i.e. from low to high. In the

area having the high hygienic condition, out of 14 samples 2 were found positive which was 14.28% of the screened individual from the respective environment and about 0.66% of the total studied population. From the medium hygienic environment, 50 out of 233 samples were positive for the infection

which was 21.45% of the respective environment and 16.66% of the total population. In the low hygienic environment, 11 out of 53 samples were found

positive which was 20.7% of the participants from the respective condition and 3.66% of the total study population as shown in Table 4.

Table 4. Seroprevalence of *T. gondii* in different hygienic environment.

Hygienic condition	Total samples (n)	Negative	Positive	Prevalence %
High	14	12	2	14.2%
Medium	233	183	50	21.4%
Low	53	42	11	20.75%
Total	300	237	63	21%

Seroprevalence of T. gondii on the basis of location

The seroprevalence of the *T. gondii* was determined on the basis of location. The participants included in the study were from rural and urban areas. In the current study 9% participants were from urban areas and 91% from rural. In the urban areas, 5 out of 27

samples were found positive for the infection. A high proportion of the studied population was found positive from rural areas. Out of 273 participants, 58 were detected positive for the parasite in the respective area as shown in Table 5.

Table 5. Seroprevalence of *T. gondii* in different locations.

Location	Total samples (n)	Negative	Positive	Prevalence %
Urban	27	22	5	18.51%
Rural	273	215	58	21.24%
Total	300	237	63	21%

Seroprevalence of T. gondii on the basis of residence

The seroprevalence of *T. gondii* was determined on the basis of residence. 84 (28%) participants were living in the brick houses and 216 (72%) were living in muddy houses. 12 (14.2%) out of 84 participants

living in the brick houses were found positive. 51 (23.36%) out of 216 participants living in the muddy houses were detected positive for the infection. The results are shown in Table 6.

Table 6. Seroprevalence of *T. gondii* on the basis of residence type.

Residence type	Total samples (n)	Negative	Positive	Prevalence %
Bricks House	84	72	12	14.2%
Muddy House	216	165	51	23.36%
Total	300	237	63	21%

Seroprevalence of T. gondii on the basis of routine contact with animals

In the current study, the seroprevalence of the infection was determined on the basis of participant's routine contact with animals. The prevalence was higher in the individuals having regular contact with animals which included cow, goat, buffalo, sheep,

donkey, and cats. 252 participants were having routine contact with such animals and out of those individuals, 58 (23.01%) were having the infection. While 48 individuals were not having any contact and out of those participants, 5 (10.4%) were detected positive for the parasite as shown in Table 7.

Table 7. Seroprevalence of *T. gondii* on the basis of routine contact with domestic.

Contact with animals	Total samples (n)	Negative	Positive	Prevalence %
Yes	252	194	58	23.01%
No	48	43	5	10.4%
Total	300	237	63	21%

Seroprevalence of *T. gondii* on the basis of marital status

The seroprevalence of the *T. gondii* was also calculated on the basis of marital status. In the current study, 233 participants were married and the

prevalence was estimated to be 57 (24.46%). While 67 samples were from unmarried individuals, in which 6 (8.9%) were detected positive for the infection as shown in Table 8.

Table 8. Seroprevalence of *T. gondii* on the basis of marital status.

Marital Status	Total samples (n)	Negative	Positive	Prevalence %
Married	233	176	57	24.46%
Unmarried	67	61	6	8.9%
Total	300	237	63	21%

Discussion

The prevalence of *T. gondii* has been used as an indicator for the endemicity of the parasite in humans and animals (Sousa, Saenz *et al.* 1988, Ades, Parker *et al.* 1993, Jones, Kruszon-Moran *et al.* 2001, Studeníčová, Benčaiová *et al.* 2006). Previously the epidemiology of the parasite has been investigated in several countries but significant knowledge is not widely available on the occurrence and transmission of the infection. In the current study, the objective was to determine the seroprevalence of *T. gondii* in the male population from district Charsadda KPK.

Out of 300 samples, 63 (21%) were found positive for the parasite and the remaining were negative. The current study shows higher frequency of the infection as compared to the other studies reported from various areas of Pakistan. In 1991, 17.4% prevalence was reported (Sadaruddin, Agha *et al.* 1991) and 17.5% reported in 2017 from Khyber Pakhtunkhwa (KP) (Shah, Zahid *et al.* 2017). However the current prevalence is less as compared to the study reported in 2007 with prevalence 40.9% (Ally and Idris 2004), 29.45% in 2012 (Tasawar, Aziz *et al.* 2012), 42% in 2014 (Hayat, Tasawar *et al.* 2014), and 58.5% prevalence in 2017 in the general population of Pakistan (Arshad, Cheema *et al.* 2017).

The prevalence and transmission of the parasite depend on several factors including hygienic condition, nutritional habits, socio-economic status, immunity, consumption of raw or undercooked meat and exposure to contaminated soil.

Among the various age groups, a higher prevalence of infection was observed in the age group of 46-55 years with 35.41% frequency. However, a lower prevalence was observed in the age group 15-25 years. From the results, it was concluded that the infection incidence rate increases with age which is significant because of host immunity. *T. gondii* infection is high in regions having poor hygienic condition where people consume undercooked meat, unwashed vegetables, and fruits. Comparatively the infection is more prevalent in rural areas as compared to urban areas as shown in Table 5. The high prevalence observed in the participants from rural areas is significant because of lack of education and unhygienic environmental condition. People having routine contact with cats, dogs, and other domestic animals or have direct contact with the soil possess a greater chance of having the infection. Toxoplasmosis is more common in those areas where people drink municipal water.

Conclusion

T. gondii still poses a major threat to the general population of Pakistan. The infection is still highly prevalent as concluded from the results. The infection rate is higher among the population having poor hygiene, regular contact with animals and in the regions where there is a lack of awareness and basic knowledge about the infection, possible transmission routes, and healthy routine habits. Efforts should be taken to assure the implementation of food safety regulations and control procedures. Examination of the pregnant women and children should be

conducted for the detection of antibodies which is significant for infection diagnosis and prescribing proper treatment. Proper animal welfare and husbandry practices should be set up and promoted for livestock, which will have a significant impact on the reduction of human infection. If such regulations and strategies are properly implemented, it will substantially reduce the burden and prevalence of human and animal infections with *T. gondii* in Pakistan.

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Conflict of interest

The authors of this paper have no conflict of interest.

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