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Seroprevalence of Human Toxoplasmosis in Southern Punjab, Pakistan

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ABSTRACT

Toxoplasmosis is a parasitic infection caused by *Toxoplasma gondii* which affects various species of mammals and birds. The infection produces a wide range of clinical syndromes in human beings. This disease causes blindness and mental retardation in congenitally infected children. The objective of present study was to determine the seroprevalence of *Toxoplasma (T.) gondii* in southern Punjab. Five hundred and fifty random samples of humans were collected. Out of 550 hosts, 162 were infected with *T. gondii*. The overall seroprevalence of *T. gondii* was 29.45%. Relationship between sex and *T. gondii* revealed a significant difference between genders. The males showed higher (35.89%) seroprevalence as compared to females (25.9%). Relationship between age and *T. gondii* showed that the parasite had highest ($P < 0.05$) prevalence (57.14%) in age group of 1-10 years and lowest (29.41%) in age group of 61-70 years. In conclusion, the overall seroprevalence rate of toxoplasmosis was 29.45% in southern areas of Punjab. Its prevalence decreases as the age of host increases and gender showed significant difference with higher seroprevalence in men as compared to women.

INTRODUCTION

Toxoplasma (T.) gondii is one of the common zoonotic parasites with worldwide distribution. It leads to abortions and neonatal problems in humans and exerts negatively impact on livestock production. Toxoplasmic encephalitis has been reported as a cause of death in immune-compromised individuals with AIDS (Dubey and Jones, 2008).

Serological surveys have been done in different parts of the world and show that more than one third of human population has antibodies against *T. gondii* (Ashburn, 1992). The seroprevalence estimates for human populations vary greatly in different countries, cultural habits within one country and among different ethnic groups living in the same area (Dubey and Jones, 2008). Around 29.5% of the adult population in district Dera Ghazi Khan, Punjab, Pakistan has serological evidence of infection (Tasawar et al., 2011). The vast majority of these human infections are either asymptomatic or result in only mild clinical disease. Thus, over the past 3 decades antibodies to *T. gondii* have been detected from 0 to 100% of individuals in various adult human populations (Flegr et al., 2003). It has been reported that the prevalence is higher in warm

and humid areas (Coelho et al., 2003). Serological surveys indicate that about 80% of all primary infections are asymptomatic, due to the effectiveness of the immune system. The tissue parasitism during the proliferative phase may occur without symptoms.

A marginal of healthy persons infected with *T. gondii* after birth develop symptoms, which are usually mild and include symptoms such as fever, malaise and lymphadenopathy (Remington et al., 2006). However, in rare cases, healthy humans develop severe and even lethal disease including pulmonary and multivisceral involvement, possibly from more virulent types of the organism (Demar et al., 2007). In addition, upto 2% of healthy persons infected with *T. gondii* develop ocular disease usually retinochoroiditis (Holland, 2003).

Various studies have been carried out on the prevalence of *T. gondii* in different parts of the world (Bahia-Oliveira et al., 2003; Basso et al., 2005; Dubey and Jones, 2008) and in Pakistan (Sadaruddin et al., 1991; Tasawar et al., 2011). An intense deficiency was felt for human toxoplasmosis in Southern Punjab, Pakistan. The current study was an effort to determine the overall prevalence relationship between age, sex and human toxoplasmosis.

MATERIALS AND METHODS

The present study was conducted to evaluate the seroprevalence of *T. gondii* in humans in southern Punjab, Pakistan. During the study, samples were collected from 550 randomly selected humans, including one hundred and fifty from District Rajanpur, one hundred from District Bahawalnagar and the remaining three hundred from Shujabad and Nishtar Medical Institute, District Multan. The age and sex of the human were recorded. Three to five ml of blood was drawn from a superficial vein of upper limb (fore arm) with the help of disposable syringe. It was then transferred into sterile screw capped tube slowly and carefully to avoid haemolysis. It was allowed to clot then centrifuged at 3000 rpm for 15 minutes. Serum was separated and transferred with the help of micropipette to another sterile serum cups and stored at -20°C until processed for analysis. The commercial “Latex Agglutination Kit” (Antec Diagnostic Product, UK) was used for the analysis of specific immunoglobulin (IgG) antibodies of *T. gondii*.

Procedure

The test procedure was carried out according to the method describe by manufacturer. The serum and reagents were brought to the room temperature. All samples were diluted at the rate of 1:20 by using LAT buffer and mixed well. A drop of each diluted serum (40µl) was placed into each well of the test slide followed by addition of a drop of latex reagent and mixed well. The presence and absence of agglutination was observed within the period no longer than four minutes. The positive sera indicated the clear agglutination, while in negative sera no agglutination was observed. The results were expressed in percentages and the values between different groups were compared by Chi Square test (Chaudhery and Kamal, 2000).

RESULTS AND DISCUSSION

Overall seroprevalence of *Toxoplasma gondii* in humans

The present study showed that the overall seroprevalence of *T. gondii* was 29.45%. Many reports from various part of the world confirm to the prevalence of toxoplasmosis in humans. The present prevalence was much higher than the 17.4% reported for humans from Islamabad, Pakistan (Sadaruddin et al., 1991) and 7.1% in Iran (Rostami et al., 2006). However, it is approximately the same as the 30.9% reported for humans in Bombay, India (Meisheri et al., 1997) and 24.2% in Slovakia (Studenicovaa et al., 2006). The higher (49.8%) prevalence rate was reported by Chacin-Bonilla et al. (2003) in Western

Venezuela. Avelino et al. (2004) detected 65.8% prevalence of human toxoplasmosis in Goiania-GO, Brazil. Singh and Pandit (2004); Sharif et al. (2007) showed overall IgG seroprevalence rate of human toxoplasmosis as 45% in India and 77.4% in Iran respectively.

The results of all these studies showed that prevalence of *T. gondii* may be high in those communities, who eat raw or undercooked meat, unwashed vegetables or fruits (de Moura et al., 2006), interaction with cats and dogs or other animals, contact with contaminated soil (Etheredge et al., 2004). Increased seroprevalence of *T. gondii* in different regions of the world can also be linked with consumption of municipal water (Ertug et al., 2005).

Relationship between sex and *Toxoplasma gondii* in humans

Sex of the hosts play an important role in the prevalence of toxoplasmosis, male hosts showed significantly ($P<0.05$) higher prevalence 35.89% as compared to female hosts 25.9% (Table 1). Moschen et al. (1991) detected a slight predominance among males (18.2% vs. 17.5% in females) in Italy. Yaneza and Kumari (1993) showed 35.6% prevalence of *T. gondii* in males and 1.9% prevalence in females in Saudi Arabia. Meisheri et al. (1997) tested sera for toxoplasmosis in Bombay, India and reported the 34% seroprevalence in men and 26.2% in women. Lee et al. (2000) reported positive rates of *T. gondii* in males and females as 7.2% and 6.0%, respectively from rural area Okcheon-gun, Korea. Sharif et al. (2007) reported higher (80%) seroprevalence in males as compared to females (77.6%) among the inhabitants of rehabilitation centers of Northern Iran.

Table 1: Relationship between sex and *T. gondii* in humans in Southern Punjab, Pakistan

Gender	Hosts Examined	Hosts Infected	Prevalence (%age)
Male Hosts	195	70	35.89
Female Hosts	355	92	25.9

$P<0.05$

The above comparison shows that the seroprevalence of toxoplasmosis is higher in males as compared to females. This could be due to different occupational, social activities and less immunity to *T. gondii* in males as compared to females. Levels of immunoglobulin, including IgG, IgM, IgA are greater in females than in males (Al-Qureshi, 2004). The literature generally indicates that the females are more resistance to parasitic infections than males because of the gender-associated differences in exposure and testosterone immunosuppressive properties (Morales-Montor et al., 2004).

Table 2: Relationship between age and seroprevalence of *Toxoplasma gondii* in Southern Punjab, Pakistan

Age groups (years)								
1-10	11-20	21-30	31-40	41-50	51-60	61-70	71-80	81-90
n=28	n=150	n=203	n=77	n=41	n=21	n=17	n=7	n=6
16	46	36	27	21	6	5	3	2
(57.14%)	(30.66%)	(17.73%)	(35.06%)	(51.21%)	(28.57%)	(29.41%)	(42.85%)	(33.33%)

P<0.05

Relationship between age and *Toxoplasma gondii* in humans

During the present study highest 57.14% positive rate of *T. gondii* was seen in the age group of 1-10 years and lowest 29.41% prevalence was recorded in the age group of 51-60 years (Table 2). Yaneza and Kumari (1993) reported highest rate (61%) of seropositivity in the age group of 21-30 years from Saudi Arabia. Eighty percent prevalence was recorded in 30-45 years age group of Ontario Veterinary Medical Staff, Canada (Shuhaiber et al., 2003). Ally and Idris (2004) reported 60% seroprevalence of toxoplasmosis in age groups 21-40 years in General Hospital Karachi. Studenicovaa et al. (2006) reported 48% prevalence of *T. gondii* in the age group of 10 years in Slovakia. Sharif et al. (2007) reported the highest (39.2%) seropositive rate of *T. gondii* in age group of 19 years in Iran.

The results of these studies showed that the prevalence of *T. gondii* was higher in younger age. Higher prevalence in young people may be due to association with pets, poor sanitary habits and low immunity against this parasite (Sadaruddin et al., 1991; Etheredge et al., 2004; Jones et al., 2008).

Relationship between different locality and *Toxoplasma gondii* in humans

The seroprevalence of human toxoplasmosis was significantly (P<0.05) low 16.33% in Multan as compared to 44.33% in Rajanpur and 51.0% in Bahawalnagar (Table 3). It has been recognized that toxoplasmosis has global distribution in humans and other warm blooded vertebrates (Young-Ha et al., 2000). The infection rate may differ from one country to another and also in different areas of the same country (Dubey and Jones, 2008). One half billion human infection is a conservative estimate. Throughout the world, the seroprevalence of toxoplasmosis in humans show great variation, ranging almost from 0 to 100% (Flegr et al., 2003) and in Europe, from 2 to 92% (Tenter et al., 2000). Previous studies of toxoplasmosis in Iran (Sharif et al., 2007), Slovakia (Carmen et al., 2006) and the USA (Dubey and Jones, 2008) have also showed a lower prevalence among individuals living in a cold or dry climate.

This difference may be explained by the influence of climate on the survival of *Toxoplasma* oocysts in the environment (Ashburn, 1992). In addition different eating habits and differences in husbandry of domestic animals may also influence exposure to infection.

Table 3: Seroprevalence of *T. gondii* in humans in different locality of Southern Punjab, Pakistan

Name of Place	Hosts Examined	Hosts Infected	Prevalence (%)
Multan	300	49	16.33
Bahawal Nagar	100	51	51.0
Rajan Pur	150	62	41.33
Total	550	162	29.45

P<0.05

We observed a decreasing trend in the seroprevalence of toxoplasmosis as the age of host increases and gender showed significant difference with higher seroprevalence rate in men as compared to women.

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