Toxoplasmosis in Small Ruminants from Varied Habitats

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Abstract: Background: Toxoplasmosis is an infectious disease found all over the world in all the mammalian and avian species. We conceived the hypothesis that the infection might be affected by the various factor differing in various habitats. The present study was conducted to evaluate our hypothesis by investigating the rates if infections different flocks of small ruminants populated in various localities with varied ecological conditions in southernmost part of Punjab, Pakistan. Materials and Method: The blood samples collected from mixed farms of sheep and goats were examined by Latex Agglutination Test to detect rates of seroprevalence of anti-Toxoplasma antibodies. Statistical analyses were applied to ascertain the correlation between habitat variations and rates of toxoplasmosis in three distinct regions of study area including sand dunes of Cholistan desert, Agricultural Region and Marshy areas. Results: The total infection rates collectively found in small ruminants were 31.41%. The incidence rates of Toxoplasma infection in small ruminants were 38.1%, 32.1% and 14.5% in animals reared in Agricultural Region, Riverside region and Sand Dunes region of Cholistan correspondingly. Conclusions: The results showed the varied rates of toxoplasmosis in small ruminants in various habitats in the study area. Furthermore, the overall higher occurrence of toxoplasmosis in small ruminants underlined the threat of zoonotic transmission of infection to human.

Keywords: Toxoplasmosis, Small Ruminants, Habitat Variations

1. Introduction

The Domestic sheep and goats are the vital source of milk, meat, fibers and pelts in almost all the countries of the world. These livestock animals are vulnerable to different parasitic diseases (Gebremedhin et al., 2013) including toxoplasmosis (Othman and Al-Azuheir, 2014) that causes a range of reproductive problems such as prenatal and postnatal mortality, still births or abortions (Edwards and Dubey, 2013) leading to the economic losses and threats to human health via zoonotic transmission (Othman and Al-Azuheir, 2014). The livestock animals in general and small ruminants in particular are exposed to a variety of infectious and noninfectious diseases of parasitic origin (Akhter and Arshad, 2006; Siddiki et al., 2010) leading to the reduction in productivity (Haileleul, 2002). Parasitic infections pose threats to health and limit the productivity due to the associated morbidity and mortality (Nwosuet al., 2007) or due to early death in the embryonic life, mummification, abortion, stillbirth (Panadero et al., 2010), and in some cases the neonatal or postnatal death (Edwards and Dubey, 2013). These infections have become a serious universal threat for livestock (Lashari and Tasawar, 2011).

One of these infections is toxoplasmosis caused by *Toxoplasma gondii* (hereafter referred as *T. gondii* or/and *Toxoplasma*) belonging to class Coccidia of phylum Apicomplexa (Ferguson, 2002; Kopečna et al., 2006). *T. gondii* is widespread in distribution and can be considered as one of the most successful protozoan parasites (Dorskaya et al., 2006) that can inflict serious diseases in all the endothermic animals (Dorskaya et al., 2006; Dubey, 2009) such as mammals including small ruminants and humans (Aspinall et al., 2002). Since *Toxoplasma* is transmitted through zoonosis particularly from sheep and goats (Sevgili et al., 2005), there is dire need to screen the animals whose meat is used by human as food so that the human health is ensured (Hill and Dubey, 2002; Lhafi et al., 2004).

Toxoplasmosis is deleterious in terms of both economy of a country and health of its people (Hill et al., 2005) and approximately thirty three percent of animals and human population of the world has been estimated to be infected with *T. gondii* at an average (Sensini, 2006) ranging between 1 and 99% rates of infections (Olivier et al., 2007). The open-air meat markets are considered as a potential source of infestation of human by *T. gondii*. While being transported from slaughter houses, the meat is by far contaminated with

oocysts of *T. gondii* hence resulting as a risk factor of zoonosis to human beings. The incidence of *T. gondii* seroprevalence in different species of animals has been studied in many countries of the world with results varying from country to country, region to region, herd to herd and season to season. Sometimes even these results vary with variation of methodology utilized on the same herd (Yu et al., 2007). The open-air markets are common in Rahim Yar Khan and Rajan Pur regions of southern Punjab, Pakistan and create a potential source of pathogen contamination as the stray cats and dogs are usually found around the markets which, being a natural reservoir of *T. gondii*, add many millions of oocysts of parasite through their excreta just within a couple of week after getting infected (Dabritz et al., 2007). Oocysts of *T. gondii* exhibit a characteristic resistance against a broad spectrum of disinfecting agents due to structural stability principally in warm and humid regions of the world, nevertheless having poor survival in arid and cold climatic conditions (Rorman et al., 2006).

Toxoplasmosis infection has been established as a major economic threat for sheep and goat farming industry particularly due to abortion all over the world (Buxton et al., 2007; Innes et al., 2009). This disease may also cause the monetary losses in terms of the reproductive disorders such as abortions, stillbirth, or weakness of neonates that results in death (Bueno et al., 2004; Soares et al., 2009) or adult mortality, reduced productivity in small ruminants with lower quality of wool and leather (Parthiban et al., 2005). According to the findings of Kijlstra and Jongert (2009), *T. gondii* is a food hazard as well.

As the survival of *T. gondii* is affected by environmental factors (Chacin-Bonilla and Sanchez-Chavez, 2000; Hill et al., 2005), we chose to conduct the current study to ascertain the variations in the rates of infections in the animals populated the remote areas of Pakistan. The present study is based on the research conducted to ascertain the association between toxoplasmosis in small ruminants' flocks reared in different habitat of Southern Punjab, Pakistan.

2. Materials and Methods

A total of 1200 blood samples were randomly collected from domestic small ruminants belonging to different localities in Cholistan Sand Dunes region, Agricultural region of district Rahim Yar Khan and Riverside (also called Kacha) Region in district Rajan Pur along the eastern bank of River Indus. Three to five ml of blood was collected from each animal's jugular vein in a vacuum tube. Each sample was allowed for 1 hour time to coagulate and afterwards applied 3000 RPM centrifugation for 10-15 minute so that serum could be decanted from the sample. The serological assay to detect the occurrence of anti-Toxoplasma antibodies (IgG) was carried out by using "Toxoplasmosis Latex" manufactured by "ANTEC DIAGNOSTIC PRODUCTS-UK". The results were analyzed statistically by applying the statistical tests Chi-square test to compute the correlation via Pearson's test through SSPS version 20.

3. Results and Discussion

In the present study, the relationship between habitat variation and occurrence of toxoplasmosis was analyzed and significantly ($P=0.000$) varied rates of toxoplasmosis infection were found in three different regions of our study area (Table 1). The results revealed that amongst total 556 small ruminants sampled from the agricultural areas region 212 were found positive for anti-Toxoplasma antibodies with the highest rate of toxoplasmosis (38.1%) followed by the animals populated in riverside areas region where, out of 404 total sampled animals 130 were positive for anti-Toxoplasma antibodies showing the infection rate at level of (32.1%) and with least rate of toxoplasmosis infection were found in the small ruminants reared in Cholistan desert region 35/240 (14.5%) as shown in Table 1.

Table 1. Relationship between toxoplasmosis and habitat variations.

Region	Animals examined	Animals positive	Prevalence (%)
Desert Areas	240	35	14.58
Agricultural Region	556	212	38.12
Riverside Region	404	130	32.17
Total	1200	377	31.41

Chi-Square = 43.2994 P-Value = 0.000

The results demonstrated the significant (P -Value= 0.000) regional variations in prevalence of Toxoplasma infection consistent with (Yang et al., 2013) who reported same picture in China and (Sechi et al., 2013) whose results demonstrated the association between toxoplasmosis and still water sources contaminated by cats' access to water consumed by small ruminants one of the important determinants in three different regions in the present study area.

These variations in infection rates probably could also be related with the humidity being highest in the habitat of small ruminants' flocks populated in the riverside areas in an agreement with Rorman et al. (2006) that the oocysts of *T. gondii* exhibit a strong structural stability predominantly in warm and humid regions of the world. It is widely accepted view that the oocysts of Toxoplasma have a poor survival in arid and cold climatic conditions (Van der Puijeet al., 2000; Rorman et al., 2006).

The higher rate of toxoplasmosis (38.1%) (Table 1) in the densely populated Agricultural Region inhabiting animal as compared with the infection rates in the animals reared in desert areas (14.5%) (Table 1) can be attributed to the presence of higher number of cats in the densely populated localities which can boost the number of oocysts in soil (Petersen and Schmidt, 2003) at a rate of many millions just by single defecation (Lopes et al., 2008) thus causing the contamination of the surrounding habitat (Silva et al., 2001) as is evident from the current study area results.

The ecological factors must be taken into account for the variation in the infection rates amongst the small ruminants reared in different habitats of study area (Dubey et al., 2004; Meerburg and Kijlstra, 2009). The prevalence rates of toxoplasmosis observed in three regions of our study area

were in agreement with (Yu et al., 2007) who reported the association between occurrences of *T. gondii* infection and environmental factors. The similar agreement was found between our findings and (Zewdu et al., 2013).

The rapid sugar industry in Rahim Yar Khan has encouraged the replacement of cotton with Sugarcane and rice crops which increased the humidity in the region suggesting the better chances for the growth of *T. gondii* in small ruminants and perhaps in the other animal species as well. On the other hand, the animals reared in Cholistan continuously face the stress of the adversely hot summer season along with the scarcity of water and natural vegetation, the mere source of fodder. This is the reason that the nomads of Cholistan desert migrate in the Agricultural areas Region including Sadiqabad, Khan Pur and Liaquet Pur in search of the fodder for their animals particularly on the onset of drought in the dog days of summer season every year. Perhaps during the immigration of the nomad tribes of Cholistan, their small ruminants contract the *Toxoplasma* infection from the highly infected animals of Agricultural localities.

Furthermore, the Dhands (the seasonal lakes formed by accumulation of rain water) in Cholistan desert, the transient resorts for Siberian migratory birds en route the lakes in the southern limits of Pakistan, are also responsible for the

horizontal transmission of *T. gondii* infection (Cabezon et al., 2011) because, we suspect, the wild cats after feeding on the migratory birds' corps must shed the oocysts in the environment (Amany et al., 2012) which might make their way to the digestive tract of the small ruminants of Cholistan. The stagnant water (Ertug et al., 2005; Sechiet al., 2013) of these Dhands might also be the source of infection as it is shared for drinking by birds as well as small ruminant species simultaneously. This hypothesis is supported by some studies reported in the near past (Taghadosi et al., 2010; Cenci-Goga et al., 2013) providing the evidences of water borne infection of toxoplasmosis.

The Riverside Region showing 32.17% infection in small ruminants exhibits the peculiar picture of humidity due to fluctuating floods in the river Indus. In this region of study area, the animals are faced with the scarcity of food in the monsoon season rainfall and some times, for many months displacement also (Ashraf et al., 2013). During the 2010 monsoon, the standing crop of cotton was devastated by flood in the river Indus. The mass migration of human population with their small ruminants occurred due to that flood thus leading to the exchange of variety of infections between animals of Rahim Yar Khan and those migrated from Kacha area region along the river Indus.

Graph 1. Prevalence of *T. gondii* infection in varied habitats

4. Conclusions

The animals reared in the urban areas with Agri-based population demonstrated the highest rates of seroprevalence of *T. gondii* (38.1%) suggesting to the hypothesis that canines must be playing a role in the multiplication of parasite. The least incidence of infections (14.5%) was noted in the Sand Dunes dwelling small ruminants perhaps due to harshly high temperature in the Cholistan desert that discouraged the sustenance of *T. gondii* while the infection rates in small ruminants 32.1% were also influenced by humid environment

produced by still water in the region.

Recommendations

- It was noticed throughout the present study that the farmers prefer to mix farming of livestock species including sheep (*Ovis aries*), goats (*Capra hircus*), buffalo (*Bos bubalis*) and cows (*Bos Taurus*) particularly where the pasture grazing adapted for feeding the animals. The standing water must be playing the role in exchange of *T. gondii* infection

among different species.

- Therefore, keeping into consideration the horizontal transmission, we suggest the screening of the other livestock species of the study area.
- We also recommend the prompt study to evaluate the infection in humans that might have transmitted by zoonosis.

Author's Contribution

The blood samples collection from sheep, serological and statistical analyses of data were performed by Saghir Ahmad under the supervision of Prof Dr Zahida Tasawar, Institute of Pure and Applied Biology, Bahauddin Zakariya University, Multan.

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