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Prevalence of toxoplasmosis in felines

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Abstract

Toxoplasmosis is the major zoonotic disease involving all warm blooded animals. Oocyst shedding is mainly seen in cats being a definitive hosts. The present study was aimed to find out the coproprevalence and seroprevalence of toxoplasmosis. Total 106 fecal samples from clinically suspected cases and apparently healthy cats were screened. The overall copro-prevalence of *toxoplasma gondii* in cats was found to be 2.83% (3/106). The prevalence was 4.91% (3/61) in animal shelters (Stray Cats) and 0% (0/45) in House hold cats (Pets). The overall sero-prevalence of toxoplasmosis by rapid test (PetX Toxo Ab) was 31.37% (16/51) with increased seropositivity in 33.33% (9/27) stray cats than pet cats 16.66% (4/24). An increase in the seroprevalence was seen as age increases. The seropositivity was higher in females than in males. Oocyst was absent in fecal samples of cats which were positive for the presence of antibodies by rapid antibody detection test kit. Regular screening by copro-microscopy is an important tool to reduce zoonotic potential of the cats.

Keywords: Toxoplasmosis, copro-prevalence, seroprevalence, Rapid antibody detection kit

1. Introduction

Toxoplasma gondii is an obligate intracellular protozoan parasite with worldwide distribution. As definitive hosts of this parasite, cats play a very important role within the life cycle of T. gondii, facilitating the genetic recombination between strains, also as environmental contamination and felines are the sole animals that pass oocysts in their faeces (Elmore et al., 2010) ^[1]. It belongs to Phylum Apicomplexa, Class Sporozoasida, Subclass Coccidiasina, Order Eimeriorina, and Family Toxoplasmatidae (Hill, 2005) ^[2]. There is just one species of genus Toxoplasma which is Toxoplasma gondii. Overall global pooled seroprevalence of Toxoplasma gondii in domestic cats reported from 1967 to 2017 was 35% which ranged between zero to 97 percent. In Australia 52% seroprevalence was seen whereas in Africa 51% was observed which was higher than other countries. In Asia, seroprevalence was lowest (27%). The highest pooled seroprevalence was noted in male domestic cats of Australia, Europe and Africa 62%, 46% and 43% respectively.(Montazeri et.al., 2020)^[3]. A serological survey in India detected 33.7% of positive cases of cats using indirect heamagglutination (Chhabra et al., 1985)^[4]. As toxoplasma gondii is one of the important pathogens affecting felines with significant zoonotic potential. As well as, there are very few studies indicating its prevalence from adjacent areas. The present study was designed to study seroprevalence and copro-prevalence of Toxoplasma gondii.

2. Material and Methods

2.1 Sample collection

The fecal samples of cats were collected in Shirwal, Mumbai and Pune area from Teaching veterinary clinical complex, Shirwal, Veterinary clinics, Animal Rescue Trusts, pet and stray cats. Samples were collected within twelve hours after defecation. Massive shedding of oocyst occurs irrespective of clinical signs hence samples were collected even without the presence of any clinical symptoms or age. Collected fecal samples were subjected to the fecal floatation method and results were noted. Blood samples were collected from the cephalic vein of cats in K3-EDTA vials and transported to the laboratory under chilling conditions. These blood samples were subjected to rapid test kits. Fecal samples were stored at -20° C till further use.

2.2 Fecal microscopy

Fecal flotation with the use of Sheather's solution was performed along with direct microscopy to visualize different gastrointestinal parasites.

The Toxoplasma IgG antibody rapid test (PetX Toxo Ab) is a test cassette to diagnose the presence of anti-Toxoplasma IgG in an animal's blood specimen in 5-10 minutes. The Toxoplasma IgG Antibody Test is based on sandwich lateral flow immunochromatographic assay. All materials, including specimens and test devices were allowed to recover to 15- 25° C before running the assay. The test device was placed horizontally. Using the capillary dropper 1 drop of the prepared specimen was placed into the sample hole "S of the test device Then 2 drops (approx. 80 µL) of the assay buffer were poured into the sample hole immediately. Test results were interpreted in 5-10 minutes. Result after 10 minutes is considered invalid.

Interpretation of results

1. IgG Positive (+): The presence of "C" line and "IgG" line, which indicates that the animal has been contracted

to Toxoplasma gondii.

2. Invalid: No coloured line appears in C zone. No matter if IgG line appears.

3. Result and discussion

3.1 Prevalence of *Toxoplasma gondii* oocysts in cat faecal sample

Out of 106 faecal samples examined, 3(2.83%) samples were found positive for *T.gondii* like oocysts by Sheather's floatation technique. (Plate 3.1 and Plate 3.2). An oocyst that resembled the morphological structure of *Toxoplasma gondi* were found in 4.91% of faecal samples collected from the animal shelters (Table 3.1). No oocysts were detected in faecal samples collected from household cats by copromicroscopy. This was probably because of feeding commercial diet and absence of hunting activities which exposes to zoonotic diseases via raw meat consumption.

Table 3.1: Pre	evalence based	on housing o	of cats
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Sr. No	Housing	No. of samples	No. of positive samples	Percent positivity (%)
1.	Animal shelters (Stray Cats)	61	3	4.91
2.	House hold cats (Pet)	45	0	0
3.	Total	106	3	2.83



Plate 3.1: Toxoplasma gondii oocyst (1)



Plate 3.2: Toxoplasma gondii oocyst (2)

Results were in accordance with Latha *et al.* (2018) ^[5] in which fecal microscopy and seroprevalence studies were done

in Thrissur, Kerala. The *T. gondii* oocyst was detected in 4.47% of 313 feline faecal samples. Similarly, Gautham, N. (2014) ^[6] studied the prevalence of Toxoplasmosis by fecal flotation and copro-PCR in Banglore. *Toxoplasma gondii* like oocyst was seen in two (1.38%) fecal samples by flotation out of total 144 samples and both samples were from animal shelters/stray cats (2.44% prevalence).

These results were comparable with Nasiru Wana (2020) [7] who used copro-microscopy to screen for the presence of T. gondii-like oocysts in 200 cat faeces, out of which 7/200 (3.5%) were detected as positive. More T. gondii-like oocysts were found in Free roaming cats (5/100, 5.0%) compared to pet cats (2/100, 2.0%). In accordance with this, Amany et al. (2012)^[8] reported that the prevalence of *T. gondii* oocysts was two per cent at Sharkia Province in Egypt by examination of 100 samples of cat faeces for the presence of T. gondii oocysts using Sheather's sugar flotation method. Also, Berger et al. (2011) ^[9] reported that only one of the cats shed T. gondii oocysts, corresponding to a T. gondii prevalence of 0.4 per cent. In contrast to this, Awobode et al. (2020) [10] identified T. gondii-like oocysts were in 21.4% (95% CI: 4.6-50.8) of the total cat faecal samples. The prevalence was 50% (95% CI: 6.7-93.3) in Akinyele community which is higher than current findings.

3.2 Prevalence of *Toxoplasma gondii* by Antibody detection kit based on housing

Out of 51 samples, 16 were found to be positive for the presence of IgG antibodies. Samples from an animal shelter/ stray cats were 27, out of them 9(33.33%) were positive and 4(16.66%) out of 24 samples were positive that were collected from household/pet cats. Prevalence study indicated that seropositivity was lower in pet cats that were fed with commercial diets compared to stray cats which were admitted in shelters for various treatments (Table 3.2)

Sr. No	Housing	No. of samples	No. of positive samples	Percent positivity (%)
1.	Animal shelters (Stray Cats)	27	9	33.33
2.	House hold cats (Pet)	24	4	16.66
3.	Total	51	16	31.37

Table 3.2: Overall prevalence of Toxoplasma gondii by Antibody detection kit

The results of the present study were in accordance with Sah et al. (2018) [11] who detected antibodies for Toxoplasma gondii using lateral flow chromatographic immunoassay (Toxo IgG/IgM Combo Rapid test®) in the Nepal region. 16 out of 44 (36.36%) cats were positive for it. Similarly, Javadi et al. (2010)^[16] detected antibodies of Toxoplasma gondii 50 cats using an enzyme-linked immunosorbent assay for IgG and IgM. Seropositivity in cats was 30% (n=15). Also, Chhabra *et al.* (1985) ^[4] did a serological survey of latent Toxoplasma prevalence study on 80 cats in northern India by the microtitre indirect haemagglutination test. Seropositivity was recorded in 33.7 per cent of 80 cats which was found to be similar to results of our study. In contrast to this, Sroka et al. (2018) [12] assessed the prevalence of Toxoplasma gondii infection in cats. In total, 208 cats (139 females and 68 males), aged 0.5-12 years (mean=2.6) from 25 localities in southwestern Poland were examined by indirect immunofluorescence assay (IFAT) to estimate the Toxoplasma gondii serological status. The positive results in

IFAT for anti-*Toxoplasma gondii IgG* and IgM antibodies were found in 143 of 208 tested cats (68.8%) which are higher than our findings.

The absence of oocyst in fecal samples of cats which were positive for the presence of antibodies by rapid test kit proved hypothesis of Dubey and Frenkel, $(1972)^{[13]}$ which depicted that, in experimental infections, cats usually stop shedding *T. gondii* oocysts by the time they seroconvert. Seropositivity in 30% cats suggests that they must have already contaminated the environment by oocyst shedding.

3.3 Age wise sero-prevalence

Age wise seroprevalence ranged from 17.64% to 50%. In kittens, 0-6 month's age range-three (17.64%) kittens were seropositive out of 17. Four (28.57%) were seropositive for the age range of 6 months to 1 year out of 14 cats. In 1-6 years and 6< years seroprevalence was 41.66% and 50% respectively i.e. five out of 12 and four out of eight respectively.

Table 3.3: Age wise sero-prevalence

Sr. No.	Age group	No. of samples	No. of positive samples	Percent positivity (%)
1.	0-6 months	17	3	17.64
2.	6 months to 1 year	14	4	28.57
3.	1-6 years	12	5	41.66
4.	6 and above	8	4	50
	Total	51	16	31.27

It was noted that seroprevalence was lower in the young age group compared to the older age group. Higher age group had more chances of exposure to *T. gondii* throughout the lifespan. (Table 3.3)

These results were in accordance with the findings of Must *et al.* (2017) ^[14] who stated that seroprevalence increased with age. Overall, 41.12% of the 1121 cats tested seropositive. Also Sroka *et al.* (2018) ^[12] assessed that the prevalence of anti-Toxoplasma antibodies was significantly greater in older cats (>1 year) (83.5%) than in younger cats (48.3%). As well

as Vollaire *et al.* (2005) ^[15] detected that seroprevalence increases with the age of cats.

3.4 Sex wise seroprevalence

Total 24 samples from males and 27 samples from females were subjected to the sandwich ELISA test kit, out of them 16 were positive. Seropositivity in male samples were 25.92%, seven out of 24 and in female samples, nine out of 27 i.e. 33.33% sero-prevalence. The seropositivity was higher in females than in males. (Table 3.4)

Table 3.4: Sex wise seroprevalence

Sr. No.	SEX	No. of samples	No. of positive samples	Percent positivity (%)
1.	Male	24	7	25.92
2.	Female	27	9	33.33
	Total	51	16	31.27

The above results were similar to the findings of Sroka *et al.* (2018) ^[12] which stated seropositivity in females was higher (74.1%) than in males (58.8%). Also, Montazeri *et.al.* (2020) found out the global pooled seroprevalence of toxoplasmosis was equal (33%, 95% CI: 29–37%) in male and female domestic cats and the difference between them was not significant. Increased seroprevalence in females can be related to sampling size and population as well as it might be related to an increase in hunting activity along with roaming to feed kittens. In contrast to this, Javadi *et al.* (2010) ^[16] detected antibodies of *Toxoplasma gondii* 50 cats using an ELISA. Fifteen (30%) of the cats were found to be seropositive and

male cats showed significantly higher antibody titres than female cats. Also Bastan and Bulent (2018) ^[17] observed a higher prevalence in male (57.1%) cats than females (42.8%). Also, Jokelainen *et al.* (2012) ^[18] noted that older cats were often found to be positive along with higher seroprevalence in cats that were fed with raw meat. Similarly, Vollaire *et al.* (2005) ^[15] noted that males had more seroprevalence than females.

4. Conclusion

The overall copro-prevalence of *toxoplasma gondii* in cats was found to be 2.83%. The overall sero-prevalence of

toxoplasmosis by rapid test (PetX Toxo Ab) was 31.37% with increased seropositivity in 33.33% of stray cats than pet cats 16.66%. An increase in the seroprevalence was seen as age increases. In 1-6 years and greater than 6 years of age, seroprevalence was 41.66% and 50%. In sex wise seroprevalence studies, 25.92% of males and 33.33% females were found to be positive for the presence of *toxoplasma gondii* specific antibodies. Oocyst was absent in fecal samples of cats which were positive for the presence of antibodies by rapid antibody detection test kit. Regular screening by copromicroscopy is an important tool to reduce zoonotic potential of the cats.

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