

The Seroprevalence of *Toxoplasma gondii* in Cats from the Kars Region, Turkey

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ABSTRACT

This study aimed to determine the seroprevalence of toxoplasmosis in indoor cats with outdoor access, in the vicinity of the Kars province. The study group consisted of 102 male and female indoor cats with outdoor access, aged 1-8 years, which were from the Kars province. Serum samples were tested by the Sabin-Feldman dye test (SFDT) for the presence of anti-*Toxoplasma gondii* antibodies. When performing the SFDT, antibody titres of 1:16 and above were considered to be positive. From the samples (n=102) tested for the presence of anti-*T. gondii* antibodies, 45 (44.1%) had an antibody titre of 1:16 and above. The distribution of seropositivity according to the location was: Kars city centre (36.8%), Akyaka (58.8%), Arpaçay (83.3%), Sarıkamış (12.5%), Selim (57.1%) and Susuz (55.6%). The differences between location seropositivity rate were found to be statistically significant (P<0.05). However, seropositivity according to age and sex was not significant (P>0.05). This study revealed first time the seropositivity the presence of *T. gondii* in cats from Kars province with high seropositivity rate of cats in this region.

Keywords: Cat; Sabin-Feldman Dye Test; *Toxoplasma gondii*.

INTRODUCTION

Toxoplasmosis is a prevalent zoonosis, which has a worldwide distribution (1). It is caused by the protozoan *Toxoplasma gondii*. The causative agent is an obligate intracellular parasite (1-4). In the life cycle of this parasite, cats can directly spread *T. gondii* in the environment (5). Thus, cats play an important role in the epidemiology of this protozoan disease (5-7). Toxoplasmosis is particularly significant, both in economic terms, due to the losses it causes in animal production as a result of prenatal death, abortion and neonatal death in sheep and goats, and in public health terms causing abortion and neonatal complications in humans (8-11).

Generally, cats are exposed to the parasite by ingesting the tissues of infected intermediate hosts. Once infected, cats shed oocysts in their feces for up to 2-3 weeks, and thereby,

contaminate the environment. Depending on the prevailing environmental conditions, oocysts may survive for several months in the environment (1, 12). Rahimi *et al.* (13) have also reported that the prevalence of toxoplasmosis is higher in temperate and humid regions, in comparison to cold and dry regions. A single cat can shed approximately 20 million oocysts into the environment in only 20 g of feces in one day (14). The transmission of *T. gondii* to humans and animals occurs as a result of the ingestion of contaminated water or food, the ingestion of raw or undercooked meat containing tissue cysts, or the transplacental passage of tachyzoites (15, 16). Major risk factors for toxoplasmosis have been identified as preying, the outdoor access of cats, and feeding on raw meat (15).

Toxoplasmosis can be diagnosed by several serological

tests, including the enzyme-linked immunosorbent assay (ELISA), Sabin-Feldman dye test (SFDT), indirect haemagglutination (IHA) and indirect fluorescent antibody test (IFAT) (3, 17, 18). Babür *et al.* (18) have reported that the SFDT is a highly sensitive method, and therefore, is used as reference test for the assessment of the sensitivity of other serological tests. This study was aimed to determine the seroprevalence of toxoplasmosis in indoor cats with outdoor access in the vicinity of the Kars province in Turkey.

MATERIALS AND METHODS

Study Site and Animals

The study was conducted after receiving approval from the Kafkas University Animal Experiments Local Ethics Committee (KAÜ-HADYEK, Investigation code: 2015/073, No 2015/095). This study was performed in Kars province in Turkey. The Kars region, located in north-eastern Turkey (43.05° E and 40.36° N), has a cold climate. The average winter temperature for the last ten years has been reported -29.6°C and lowest temperature has been measured -17.1°C in this study period. Also the lowest temperature for the last ten years has been reported to be -32.0°C (19).

The study material consisted of randomly selected 102 clinically healthy male (44.1%) and female (55.9%) indoor cats with outdoor access, aged 1-8 years from the Kars city centre and surrounding districts of Akyaka, Sarıkamış, Arpaçay, Selim and Susuz.

Collection of Samples

Five milliliter blood were collected from all animals for the extraction of sera. Blood samples were centrifuged at 3000 rpm for 10 minutes for the extraction of sera. The serum samples were stored -20°C until analyzed.

Sabin-Feldman Dye Test (SFDT)

SFDT was performed as previously described (20, 21). All serum samples were tested by the SFDT for the presence of anti-*T. gondii* antibodies. Following inactivation of complement at 56°C for 30 minutes, positive control, negative control and test sera were diluted with saline in a series of 4-fold serial dilutions from 1:4 to 1:1024. Dilutions were transferred to another tube and equal volume of activator was added. Tubes were then mixed and incubated 37°C for 50 minutes. After this process 0.025 ml buffered methylene

blue (pH=11) was added to each tube and incubated 37°C for 10 minutes. Then 0.020 ml of the sample was taken and examined under microscope (40x). When greater than 50% of the observed *T. gondii* tachyzoites remained unstained, this was considered a positive result. Antibody titer of 1:16 and over was accepted to be positive.

Statistical Analysis

Chi-square statistical analysis of the data was performed using SPSS 20.0 statistical software package. Values of $P < 0.05$ were considered to be statistically significant.

RESULTS

According to the information provided by owners of the cats included in this study all had outdoor access. Of the 102 cats tested for the presence of anti-*T. gondii* antibodies, 45 (44.1%) had an antibody titer of 1:16 and above, whilst 57 (55.9%) had an antibody titer below 1:16. The distribution of seropositivity according to the locations included in the study was as follows: Kars city centre (36.8%), Akyaka (58.8%), Arpaçay (83.3%), Sarıkamış (12.5%), Selim (57.1%) and Susuz (55.6%). The differences between the seropositivity rates of the locations were found to be statistically significant ($P < 0.05$). Of the cats determined to be seropositive, 33 had an antibody titer of 1:16, 11 had an antibody titer of 1:64 and 1 had an antibody titer of 1:256 (Table 1).

Table 1: Feline toxoplasmosis seropositivity rates and titres according to region.

Location	Number of animals	Positivity %	Titers		
			1:16	1:64	1:256
Central district	38	36.8	12	1	1
Sarıkamış	16	12.5	1	1	-
Selim	7	57.1	4	-	-
Susuz	18	55.6	4	6	-
Akyaka	17	58.8	8	2	-
Arpaçay	6	83.3	4	1	-
Total	102	44.1	33	11	1

$P=0.016$ (Between groups)

Of cats included in the study, 45 were male and 57 were female. *T. gondii* seropositivity was found in 44.4% (20/45) of the males, and 43.9% (25/57) of the female cats. No statistically significant seropositivity differences were observed between the males and females. ($P > 0.05$).

Toxoplasma gondii seropositivity according to age revealed that 41.9% of animals were >1-≤3 years of age, 37.0% were >3-≤5 years of age, and 60.0% were >5 years of age or older (Table 2). Differences in seropositivity according to age were not found to be significant ($P>0.05$).

Table 2: The seroprevalence of *Toxoplasma gondii* according to age as determined by the SFDT

Age	Number of Animals	Positive	%
>1-≤3	31	13	41.9
>3-≤5	46	17	37.0
>5	25	15	60
Total	102	45	44.1

$P=0.167$ (Between groups)

DISCUSSION

Toxoplasma gondii is a zoonotic disease which can infect humans and almost all warm-blooded animals (1). The seroprevalence of toxoplasmosis in cats is related to the geographical region, the age and outdoor access of cats, and the serological test used (22, 23). Data available on the prevalence of this protozoan disease aids in the assessment of public health and environmental contamination (6, 24, 25).

In the present study, aimed at to determine the seroprevalence of toxoplasmosis in indoor cats with outdoor access from the Kars region, seropositivity was ascertained as 44.1%. Different results have been reported in cats in previous serological research studies in Turkey. Seropositivity rates determined by the use of the SFDT have been reported as 55.5% in the Elazığ province and as 76.4% in the Niğde province (7, 26). In their study conducted in the Ankara region, Özkan *et al.* (27) reported the seropositivity rates determined by the SFDT and IFAT as 40.3% and 34.3%, respectively. Babür *et al.* (7) reported seropositivity rates of 69.8% and 78.0%, respectively, for the Kırıkkale and Sivas provinces, on the basis of IHA test results, and of 23.4% and 43.1% for the Ankara province, on the basis of SFDT results. The differences observed between these results were attributed to the number of samples collected and the geographical regions investigated. Rahimi *et al.* (13) suggested that the prevalence of toxoplasmosis is higher in temperate and humid regions,

in comparison to cold and dry regions. However, although located at a high altitude and characterized by a cold and dry climate, the Kars region presented with a relatively high toxoplasmosis prevalence of 44.1% in the present study. The results in this study are within the range of seropositivity rates reported previously in Turkey. Also in this study, seropositivity rates of *T. gondii* determined between 12.5-83.3% different regions. This difference could be attributed to the number of infected animals, cat population and the number of animals sampled at the locations.

Seropositivity rates determined in other countries also differ greatly. In Iran, a neighboring country of Turkey, Derakhshan and Mousavi (28) reported a positivity rate of 2.7% on the basis of IFAT results, whereas Sharif *et al.* (29) and Hamidinejat *et al.* (30) reported positivity rates of 40% and 54%, respectively, using the same method. Positivity rates determined by the modified agglutination test (MAT) were reported to vary between 20.5% and 54.0% (6, 22, 30-34), whilst positivity rates determined by the IFAT have been reported to vary between 2.7% and 47.6% (28, 35-37).

Varying differences in the association of *T. gondii* seropositivity according to age and sex have been observed by others in the past. The proportion of serpositivity in our study corresponds with the previous studies where no association between age was found. (7, 38). On the other hand studies also determined increased antibody as animals aged (27, 31, 32, 35). This difference could be attributed to infection pressure that animals were exposed in this region.

Although it has been reported that sex is a more important factor for the prevalence of toxoplasmosis, and that the disease is more prevalent in female cats than in male cats (36), it is generally believed that sex is statistically less important for the distribution of toxoplasmosis (7, 26, 30, 31, 35, 38). In the present study which was in agreement with this general opinion, it was ascertained that sex of the cats was a statistically insignificant factor for the seroprevalence of toxoplasmosis in the Kars region of Turkey.

In the present study, the seroprevalence of *T. gondii* in cats from the Kars region was determined as 44.1%, and this bears significance as this is to the best knowledge of the authors the first report on the prevalence of this protozoan parasite in cats from the Kars region in Turkey.

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