



Seroprevalence and Risk Factors of *Toxoplasma gondii* Infection among Cat Sitters in Korea

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Abstract: The seroprevalence of human toxoplasmosis has been increasing in Korea, and it is controversial whether cats are an important infection source or not. This study was performed to evaluate the seroprevalence of *Toxoplasma gondii* infection in a high risk group (cat sitters) and to determine the possible importance of cats as an infection source in Korea. Risk factors, including the age, sex, and diet of cat sitters, their contact experience and contact frequency with stray cats, and origin, number, and outdoor activity of their pet cats, were analyzed using structured questionnaires. A total of 673 serum samples from people who have frequent contact with cats (high risk group) and 1,114 samples from general people (low risk group) were examined for specific IgG antibodies against *T. gondii* by ELISA. The results revealed that the overall seroprevalence of *T. gondii* infection was 7.4% (n=1,787). The seroprevalence among low risk group was 8.0% (89/1,114), whereas that among high risk group was rather lower 6.4% (43/673), though this difference was statistically not significant ($P=0.211$). Among the risk factors, only the outdoor activity of pet cats was important; people having cats with outdoor activities revealed 2 times higher seroprevalence than people having cats with only indoor activities ($P=0.027$). In conclusion, the seroprevalence of *T. gondii* was not significantly different between the high risk group and low risk group, and the importance of cats as a source of infection in Korea is questionable.

Key words: *Toxoplasma gondii*, seroprevalence, cat sitter, ELISA, risk factor

Toxoplasmosis is a worldwide opportunistic zoonotic disease caused by the protozoan parasite *Toxoplasma gondii*. Almost all warm-blooded animals, including humans, can serve as the intermediate host, and its definitive host is the Felidae family, including cats [1]. Most of healthy individuals infected with *T. gondii* generally have no clinical symptoms and signs [2]. However, in immunocompromised individuals and pregnant women, the parasite can cause severe diseases, such as meningoencephalitis and abortion [2]. Humans can contract this disease through consumption of food or water contaminated with oocysts from infected cats, eating undercooked meat containing tissue cysts, or by vertical transmission from mothers to fetuses [3]. The significance of each mode of contraction needs clarification in each epizootiological and epidemiological environment and locality.

According to previous reports in Korea, the seroprevalence of *T. gondii* among general population has been reported to be around 10%; however, the prevalence tends to increase due to various possible risk factors [4]. Especially, owning 3 or more kittens in a household and frequent exposure to cat feces may be highly correlated with higher seroprevalence [5,6]. However, in Korea, there have been few studies on risk factors of *T. gondii* infection. In particular, it remains to be determined whether a high risk group, for example, cat sitters (called ‘cat-mom’ in Korean) have a higher seroprevalence compared with a low risk (control) group who do not take care of cats.

Recently, the number of stray cats has been increasing steadily in Korea. Thereby, food and water contaminated with oocysts from stray cat feces may be an important risk factor for *T. gondii* infection. However, no surveys have been conducted on *T. gondii* infection among people feeding cats in Korea. Thus, it is needed to investigate the seroprevalence of *T. gondii* infection among people who have frequent contact with cats, including domestic (pet) cats and/or stray cats. In our previous study in 2013, we examined 300 fecal samples of stray cats from all around the country and detected *T. gondii* B1 gene

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from 14 (4.7%) samples using nested-PCR [7]. In the present study, we examined the seroprevalence of *T. gondii* infection among cat sitters (high risk group) in comparison with general people (low risk group) in Korea and analyzed the importance of several risk factors using a questionnaire study.

We collected blood samples from 673 cat sitters voluntarily recruited by the Korean Organization for Protection of Cats (Seoul, Korea) in 2013. As the control group, sera of 1,114 randomly selected healthy volunteers (a few of them may have contact with cats) who visited the Korea Association of Health Promotion for health check-up were included for the serological assay. The questionnaires comprised of various items, including how to acquire their own cats, the origin of cats, and

the lifestyle of cats, in order to get information on possible routes of *T. gondii* infection. All of the blood and sera were stored at -80°C until analyzed. This study was approved by the Institutional Review Board of Seoul National University Hospital, Seoul, Korea (IRB no. C-1310-007-521). The purpose and procedures of this study were explained to all participants, and a written informed consent was obtained from each of the participants.

The process for preparing *T. gondii* lysate antigens (TLA) [4] and ELISA followed previous studies with slight modifications. Briefly, 96-well microtiter plates (Costar, Cambridge, Massachusetts, USA) coated with TLA were incubated at 4°C overnight. After washing, each well was reacted with the test serum

Table 1. Seroprevalence of *Toxoplasma gondii* among high risk group (cat sitters) in comparison with low risk group

	No. examined	No. positive (%)	Unadjusted OR	95% CI of OR	P-value
Low risk group (general people)	1,114	89 (8.0)	1.000		
High risk group (cat sitters)	673	43 (6.4)	0.786	0.539-1.146	0.211
Total	1,787	132 (7.4)			

OR, odds ratio; CI, confidence interval.

Table 2. Seroprevalence of *Toxoplasma gondii* in high risk group (cat sitters) by risk factors related with cat sitter characteristics

	No. examined	No. positive (%)	Unadjusted OR	95% CI of OR	P-value
Age					0.952
	10-19	15	1 (6.7)	1.000	
	20-29	228	17 (7.5)	0.800	0.132-4.855
	30-39	286	17 (5.9)	0.628	0.103-3.806
	40-49	99	5 (5.1)	0.563	0.081-3.911
	50-59	39	3 (7.7)	0.927	0.118-7.278
	>60	6	0 (0.0)	0.744	0.021-26.116
	Total	673	43 (6.4)		
Sex					0.943
	Male	107	7 (6.5)	1.000	
	Female	566	36 (6.4)	0.970	0.420-2.241
Diet					0.944
	Meat-based	97	7 (7.2)	1.000	
	Plant-based	63	4 (6.3)	0.872	0.244-3.109
	Mixed	508	32 (6.3)	0.864	0.370-2.019
	No answer	5	0 (0.0)		
Experience of contact with stray cats					0.499
	No	48	2 (4.2)	1.000	
	Yes	613	41 (6.7)	1.649	0.386-7.033
	No answer	12	0 (0.0)		
Contact frequency with stray cats					0.062
	<1/month	286	15 (5.2)	1.000	
	2-5/month	118	5 (4.2)	0.799	0.284-2.252
	2-5/week	94	12 (12.8)	2.645	1.190-5.875
	1/day	114	8 (7.0)	1.364	0.562-3.310
	No answer	1	1 (100.0)		

OR, odds ratio; CI, confidence interval.

samples (1:100) at 37°C for 1 hr, and horseradish peroxidase-conjugated goat anti-human IgG (1:10,000; Bethyl Laboratories, Montgomery, Texas, USA) was applied at 37°C for 1 hr. After several washings, freshly prepared o-phenylenediamine dihydrochloride (Sigma-Aldrich, St. Louis, Illinois, USA) was added, and the reaction was stopped by adding 8 N H₂SO₄. IgG antibody titers were determined at optical density of 490 nm. The risk factors of *T. gondii* infection were analyzed by multivariate correlation analysis with logistic regression (SAS system for Windows, version 9.2, SAS institute, Cary, North Carolina, USA) to find out associations with various basic components of the cat owners and their cats, including the lifestyle of cats. The level of significant difference was defined as $P < 0.05$ for each risk factor.

The results revealed that the seroprevalence of *T. gondii* infection in high risk group (6.4%, 43/673) was rather slightly lower than that of low risk group (8.0%, 89/1,114) (Table 1). However, the difference was statistically not significant ($P = 0.211$) (Table 1). The questionnaire analysis revealed little meaningful differences between the seropositivity of high risk group and low risk group in respects of cat sitter characteristics (Table 2) and cat characteristics (Table 3). The frequency of contact with cats, in particular, 2-5 times in a week, appeared to be statistically significant in cat sitters; however, the overall

P -value for the frequency of contact with cats (4 options) was 0.062, thus this risk factor could not be justified as highly significant (Table 2). A noteworthy risk factor was the outdoor activity of their cats, which showed a significant ($P = 0.027$) correlation with *T. gondii* seroprevalence (Table 3). The seroprevalence of cat sitters having a cat with outdoor activities was 2 times higher than the cat sitters having cats with indoor activities only (OR = 2.072, $P < 0.05$) (Table 3).

The risk factors of *T. gondii* infection in humans include the age and gender of cats, the way of raising cats, blood transfusion, eating habits, such as consumption of raw or undercooked meat, drinking untreated water, and eating unwashed fresh vegetables or fruits [5,8-13]. Especially, pork, lamb, and wild-game animal meat has been found to be contaminated with *T. gondii*, and such wildlife was also associated with the seroprevalence of *T. gondii* in the US [5]. Stray cats have a higher chance of exposure to *T. gondii* infection through hunting, feeding a meat, or drinking oocyst-contaminated water that may survive from months to years in soil or water in nature [14].

The results of our study indicated that, in our study setting, the seroprevalence of toxoplasmosis among cat sitters (high risk group) was not higher compared to that in general people (low risk group). Among the subjected general people, a few

Table 3. Seroprevalence of *Toxoplasma gondii* in high risk group (cat sitters) by risk factors related with their pet cats

	No. examined	No. positive (%)	Unadjusted OR	95% CI of OR	P -value
Origin of cats					0.940
No stray cat	214	13 (6.1)	1		
Stray cat	426	29 (6.8)	1.129	0.575-2.220	0.900
Unknown	15	1 (6.7)	1.104	0.135-9.063	0.971
Total	655	43 (6.4)			
Duration of living together (year)					0.678
<1	119	8 (6.7)	1		
1-2	193	16 (8.3)	1.254	0.520-3.028	0.267
3-5	166	9 (5.4)	0.795	0.298-2.125	0.533
> 5	177	10 (5.6)	0.831	0.318-2.170	0.624
No. of cats living together					0.449
1	248	18 (7.3)	1		
2-3	300	16 (5.3)	0.72	0.359-1.443	0.209
>4	105	9 (8.6)	1.198	0.520-2.761	0.378
No answer	2	0 (0.0)			
Outdoor activity of cats					0.027
No	499	27 (5.4)	1		
Yes	151	16 (10.6)	2.072	1.085-3.958	0.027 ^a
No answer	5	0 (0.0)			

OR, odds ratio; CI, confidence interval.

^aStatistically significant.

people who were rearing or who reared pet cats may have been included. According to Segye Daily Newspaper, Korea (17 December 2015), about 2.7% of Korean households were reported to have pet cats. We regret that we could not discriminate them from others and exclude such subjects. In addition, no statistically significant differences were noted in the seropositivity of people having different risk factors, including the age, gender, and type of diet of cat sitters, and their experience of contact with stray cats, as well as the origin and number of pet cats and the duration of living with cats (Tables 2, 3). Only 1 factor, i.e., rearing cats with outdoor activities was positively associated with high seropositivity of cat owners (i.e., cat sitters). This agrees to a previous report in Korea [13].

The results suggested 2 important points with respect to the epidemiological status of toxoplasmosis in Korea. One was that, although not many studies have been performed regarding *T. gondii* infections among the wildlife, it is highly suggested that this protozoan should be widely distributed among the wildlife in Korea, including stray cats. Active investigations are urgently needed to properly understand the significance of this risk factor. The other suggestion was that undercooked meat, including pork, may be a more important source of human infections compared to contact with cats. However, meat inspection targeting toxoplasmosis, particularly in pork, has not been officially executed in Korea. The importance of pork (including domestic and imported pork) as a source of human toxoplasmosis should be verified by regular monitoring and investigations.

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CONFLICT OF INTEREST

We have no conflict of interest related to this work.

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