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Soroprevalência de *Toxoplasma gondii* em gatos (*Felis catus*, Linnaeus 1758) residentes em Lima, Peru

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Abstract

The aim of this study was to determine the frequency of infection by *T. gondii* in cats, by examining serum and fecal samples from animals attended at veterinary clinics in the Metropolitan Region of Lima, Peru. We collected and analyzed 154 cat serum samples and 50 fecal samples, regardless of the age, gender or breed. In parallel with the sample collections, the owners answered an epidemiological questionnaire that investigated the following variables: age group, gender, lifestyle (confined, semi-confined or free-living animals), feeding and hunting habits. The serum and fecal samples were analyzed using indirect hemagglutination (IHA) and coproparasitological tests, respectively. IHA showed that the frequency of cat exposure to *T. gondii* was 11%. Age and gender showed no association with exposure to the parasite. Exposure among the cats was associated with hunting ($\chi^2 = 4.98$, $p = 0.016$) and feeding habits ($\chi^2 = 13.34$, $p = 0.001$): those fed with raw meat were more exposed than those fed with commercial cat food ($\chi^2 = 9.50$, $p = 0.004$) or with homemade food ($\chi^2 = 4.1$, $p = 0.027$). The frequency of cats diagnosed in the chronic phase of *T. gondii* infection was 88% (15/17). No *T. gondii* oocysts were found in any of the 50 fecal samples examined.

Keywords: *Toxoplasma gondii*, Lima, Peru, Indirect Hemagglutination, cats.

Resumo

O objetivo do presente trabalho foi determinar a frequência sorológica e coproparasitológica da toxoplasmose em gatos atendidos em clínicas veterinárias na região metropolitana de Lima, Peru. Foram analisadas 154 amostras de soros de gatos e 50 amostras de fezes de gato, independentemente da idade, gênero ou raça. Paralelamente ao ato da coleta, os proprietários responderam a um questionário epidemiológico onde foram tratadas as seguintes variáveis: faixa etária, gênero, estilo de vida (animais confinados, semiconfinados ou de vida livre), hábitos alimentares e de caça. Os soros e amostras fecais foram analisados pelos testes de hemaglutinação indireta (HAI) e coproparasitológico, respectivamente. A frequência de gatos expostos foi 11,0%, segundo HAI. As variáveis de faixa etária e sexo não mostraram associação com a exposição ao parasito. A exposição dos animais mostrou associação com hábitos de caçar ($\chi^2 = 4.98$, $p = 0.016$) e alimentação ($\chi^2 = 13.34$, $p = 0.001$), sendo aqueles alimentados com carne crua os mais expostos, quando comparados aos alimentados com ração ($\chi^2 = 9.50$, $p = 0.004$) ou com comida caseira ($\chi^2 = 4.1$, $p = 0.027$). A frequência de gatos na fase crônica da infecção por *T. gondii* foi 88% (15/17). Não foram achados oocistos de *Toxoplasma gondii* em nenhuma das 50 amostras de fezes.

Palavras-chaves: *Toxoplasma gondii*, Lima, Peru, Hemaglutinação indireta, gatos.

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The worldwide distribution of *Toxoplasma gondii* crosses political boundaries and environmental conditions, and results in worldwide distribution of toxoplasmosis. Infection with this protozoon is one of the most common parasitic infections of humans and other warm-blooded animals (DUBEY; BEATTIE, 1988).

Its life cycle is divided into a sexual stage that occurs in the epithelial cells of the small intestine of Felidae, resulting in formation of oocysts that are shed in their feces, and an asexual phase that occurs in extraintestinal tissues of mammals or birds (ACHA; SZYFRES, 2003). Over one million non-sporulated oocysts can be dispersed to the environment in cat feces (DUBEY; FRENKEL, 1972). When environmental conditions are favorable (water, wetland, average temperature of 25°C, and enough oxygen), the sporulated oocysts become infective within three days, thus explaining the high incidence of infection in tropical or subtropical areas (LEGUÍA, 2002).

The high rate of seropositivity among some feline populations can be explained by environmental contamination by oocysts, and consequently, elevated concentration of infective agents in food and water, which may reflect high rates of infection with *T. gondii* in intermediate hosts (PENA et al., 2006). However, it needs to be borne in mind that, contrary to common belief, immunocompetent cats only eliminate oocysts in their feces over the course of primary infection (3-20 days in their lives). Therefore, areas inhabited by seropositive cats tend to express lower parasite loads (DUBEY, 1994a, 1996).

The animal infection rates obtained in different countries around the world are influenced by different factors such as geographic location (higher rates in tropical zones) (GONDIM et al., 1999; MARTÍN-HERNÁNDEZ; GARCÍA-IZQUIERDO, 2003; SILVA et al., 2003), public cultural habits (ingestion of raw or undercooked meat and untreated water) (DUBEY, 1988), type of fauna, infrastructure and the degree of development of the area. In this light, serum and fecal samples from household cats were assayed to determine the frequency of exposure to *T. gondii*, using commercial hemagglutination and coproparasitological tests, respectively. This was done to test the serological and coproparasitological prevalence of feline toxoplasmosis as an indicator of parasite spreading in the Metropolitan Region of Lima, Peru.

The project was authorized by the Ethics Committee for Animal Research, Dean's Office for Research and Postgraduate Studies, Universidade Federal Fluminense, Niterói, RJ, Brazil.

The survey was conducted in the Metropolitan Region of Lima, Peru (12°02'S, 77°01'W). After owners' consent had been obtained, 154 cats, irrespective of age, breed, gender, and health status were sampled for blood and 50 of these, for feces. The survey data form was completed by including information on gender, lifestyle (confined, semi-confined or free) (MENDES-DE-ALMEIDA et al., 2005), age, diet and hunting habits. The animals were categorized as young (<1 year), adult (1-7 years) and over 7 years old (HAND et al., 2000). Diet was categorized as: (i) cat food, those that were exclusively fed with commercial diet; (ii) homemade food, those fed with homemade food or homemade food and commercial diet; or (iii) raw meat, those also fed with raw meat in addition to other sources.

Screening for antibodies was performed using the indirect hemagglutination (IHA) test (TOXO HAI-Lab, Analisa, Belo Horizonte, Brazil), following the manufacturer's instructions. Fecal samples were processed by means of the FAUST technique (FAUST et al., 1938).

A database was created, using EPI INFO 2002 (CDC, 2002), which included the identification data of each animal (sex, age, eating habits, lifestyle and hunting habits) and laboratory test results. The data were subjected to exploratory analysis (MEDRONHO et al., 2002) and later to the chi-square and Fisher Exact statistical tests (SAMPAIO, 2002), to investigate the existence of associations between the variables.

A total of 17 cats were tested positive for *T. gondii* antibodies (11%). The variables of age and gender did not show any association with exposure. In the Metropolitan Region of Lima, exposure of animals was associated with hunting habits ($\chi^2 = 4.98$, $p = 0.016$) and feeding habits ($\chi^2 = 13.34$, $p = 0.001$). Cats that were fed with raw meat were more exposed than those fed with commercial food ($\chi^2 = 9.50$, $p = 0.004$) or homemade food ($\chi^2 = 4.1$, $p = 0.027$) (Table 1).

The majority of the exposed cats (88%; 15/17) only showed markers of past exposure (IgG). Interestingly, the two animals that were identified with acute-phase markers (2/17) were more than seven years old.

Since this parasitic species is undemanding with regard to environmental conditions, it is considered to persist with greater frequency in warm humid areas (MARTÍN-HERNÁNDEZ;

Table 1. Absolute and relative frequencies of antibodies to *Toxoplasma gondii* according to the indirect hemagglutination (IHA) test, distributed according to the variables of age, gender, lifestyle, diet and habit of hunting, among cats in the Metropolitan Region of Lima, Peru, 2011.

Variables	Lima		
	Total	Positive	% ± IC
	154	17	11.0 ± 4.9
Age			
<1 year	49	3	6.1 ± 6.7
1 year-7 years	43	6	13.9 ± 10.3
>7 years	62	8	12.9 ± 8.3
Gender			
Male	79	11	13.9 ± 7.6
Female	75	6	8.0 ± 6.1
Lifestyle			
Confined	107	9	8.4 ± 5.2
Semi-confined or free	47	8	17.0 ± 10.7
Feeding			
Commercial food	111	9	8.1 ^a ± 5.0
Homemade food	35	4	11.4 ^a ± 10.5
Raw food	8	4	50.0 ^b ± 34.6
Hunting			
Yes	42	9	21.4 ^a ± 12.4
No	112	8	71.4 ^b ± 4.7

Different letters in columns indicate 95% significance, in relation to the same variable.

GARCÍA-IZQUIERDO, 2003). These are ideal conditions for maintaining the viability of the oocysts. A previous study (DUBEY, 1994a) demonstrated that oocysts could remain viable at temperatures between 10 and 25°C, which makes it clear that the Metropolitan Region of Lima has ideal conditions for development and survival of the parasite.

This parasitic species shows high adaptability to its environment. These adaptability skills are demonstrated by the fact that it has been reported on every continent of the planet, except for certain Pacific islands where there are no definitive hosts (WALLACE, 1969). Thus, cats are important sources for maintenance of the parasite species in urban areas, regardless of region, since the species is found worldwide.

The sex and age of the hosts did not have any influence on exposure rates, which was expected (DEEB et al., 1985; GARCIA et al., 1999; OVALLE et al., 2000; GAUSS et al., 2003). Nonetheless, it has been suggested that males are more exposed than females (MIRÓ et al., 2004) because they use greater areas for roaming, as long as they remain free to roam outdoors without human supervision (SMITH et al., 1992). The cats that presented the acute phase of infection were over seven years of age, and therefore it can be suggested that cats will become more susceptible to disease through variations in hygiene habits or diet, presence of parallel infection with certain retroviruses or development of tumors that induce immune suppression. In this manner, parasite reactivation (VENTURINI et al., 1997) sometime later in life is promoted, with consequent production of immunoglobulin M. In our study, parasite reactivation seems to be the most likely event, since we could not find any fecal samples containing oocysts, even in animals positive for IgM in serum samples.

It was noted that cats that had the habit of hunting and eating raw meat had greater exposure to this parasite. It is likely that free access to raw meat increases exposure to *T. gondii* because of survival of these parasites inside chilled and refrigerated meat, which is the local way to preserve beef and beef viscera products for trade (ESPINO, 2006).

Some owners stated that although their cats were outdoor cats, they would not hunt at all because they lacked this habit. We were not able to assess the accuracy of such claims, meaning that these could be mistaken, given that hunting has been widely associated with exposure to *T. gondii* (VENTURINI et al., 1997; LUCAS et al., 1999; LOPES et al., 2008; LÓPEZ et al., 2011). Therefore, it can be inferred that the habit of hunting was associated with infection by *T. gondii*, and consequently, veterinarians must guide people responsible for these animals to keep them well fed and prevent their access to prey. Moreover, those responsible for cats that do hunt should be instructed to wash the litter boxes daily, to prevent development of infectious parasitic forms within feces deposited in these litter boxes and thus inhibit the parasitic cycle.

Despite the observation that 2/17 (11.7%) of all the examined animals were in the acute phase of infection by *T. gondii*, as demonstrated by IgM levels, we were not able to find any oocysts in their stool samples. This strongly suggests that those IgM levels reflected reactivation of parasitic forms within these hosts. The results from this study suggest that despite all efforts aiming to promote healthy living habits among domestic cats and other species in urban environments, different parasitic species

can still be transmitted between cats and other hosts in these environments. Thus, promoting awareness of such parasitic diseases and encouraging the development of surveillance mechanisms must be strengthened, especially regarding parasitic species such as *T. gondii*. It was observed that this parasite has great adaptability skills, particularly with regard to environmental conditions. Most cats receiving human care have not been exposed to the parasite before, and therefore they might be able to eliminate large amounts of oocysts if they become infected, thereby increasing the risk of household transmission. It is worth noting that animals that have been exposed to this parasite and then become reinfected usually do not eliminate oocysts at all. Even if they do so, it is only done in small amounts in their faeces (DUBEY, 1994b).

The seroprevalence of *Toxoplasma gondii* in cats living in the Metropolitan Region of Lima was 11%. The gender of the hosts did not influence rate of exposure to the parasitic infection in this study.

The exposure to *T. gondii* was associated with hunting and feeding habits: those fed with raw meat were more exposed than those fed with commercial cat food or homemade food.

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