

Prevalence of *Toxocara canis* and other endoparasites in client-owned dogs from Cluj county, Romania

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Abstract. *Toxocara canis* is a worldwide endoparasite in dogs with a high zoonotic potential and one of the main cause of human toxocarosis. The aim of the study was to establish epidemiological aspects of toxocarosis in client-owned dogs from Cluj County. Feces samples were collected from 155 dogs and analyzed by flotation method using sodium chloride saturated solution.

The overall prevalence of parasites was 67.1% (104/155). *Toxocara canis* was the most frequent parasite found (54/155; 34.8%), in single (31/155; 20%) or mixed (23/155; 14.8%) infections. The most prevalent mixed infection was between *T. canis* and *Isospora* spp. (8.4%; 13/155). The risk factors that influenced the prevalence of toxocarosis were age (puppies) and absence of deworming ($p < 0,005$).

Other parasites found were *Trichocephalus vulpis* 20.0% (31/155), *Isospora* spp. 15.5% (24/155), *Ancylostoma caninum/Uncinaria stenocephala* 10.4% (16/155), *Giardia duodenalis* 7.1% (11/155) and less frequent (1/155) *Toxascaris leonina*, *Capillaria aerophila* and *Dipylidium caninum*.

In conclusion, the high prevalence and asymptomatic infection with *Toxocara canis* and other endoparasites in dogs, but especially with those of zoonotic risk (toxocarosis, giardiasis, ancylostomiasis and dipylidiasis) represent a concern of animal and public health and require a proper management of efficient therapeutic and prophylactic measures.

Keywords: *Toxocara canis*; Zoonosis; Risk factor; Endoparasites.

Received 08.11.2016. Accepted 10.12.2016.

Introduction

In recent years, there have been an exponential increase of number of dogs and cats as companion animals due to the interest of

people in human-animal interactions and their emotional, psychological and social impact (Paul et al., 2010), especially in Romania. *Toxocara canis* is one of the most frequent parasite in dogs with a prevalence between

3.5% to 34% in Europe (Overgaauw and van Knapen, 2013) and researches are focused on its importance from both veterinary and public health aspects.

Adult worms of *T. canis* are localized in the small intestines of canids and females are able to lay eggs that pass in the environment through feces (Anderson, 2009). Patent infections usually happen in young dogs due to hepato-tracheal route of migration and may contribute to environmental contamination. Because of the development of immunological responses, adults do not shed *Toxocara* eggs as somatic migration takes place, but in different physiological perturbations, this can also be possible (Nijse et al., 2016).

Dogs can become infected with *Toxocara* through ingestion of embryonated eggs, prenatal infection from pregnant bitches, transmammary route or oral infection by consuming paratenic hosts (Schnieder et al., 2011). Humans and other vertebrates or non-vertebrates can accidentally become paratenic hosts by ingesting embryonated eggs from the environment or through contaminated fruits and vegetables Glickman and Shofer, 1987), but also by consuming raw meat that contain somatic larvae in tissues (Taira et al., 2004). In humans, larvae of *T. canis* stop from evolving, but encysts in different body tissues and causes four major syndromes: larva migrans visceralis (VLM), neurological toxocariasis (NT), larva migrans ocularis (OLM) and covert toxocariasis (Magnaval et al., 2001; Vidal et al., 2003).

In order to control canine toxocarosis, but also its consequences in public health, guidelines regarding the deworming of dogs have been developed by the European Scientific Counsel Companion Animal Parasites (ESCCAP) especially in dogs with high risk like kennel dogs, puppies or geriatric individuals, dogs that live outdoor, stray and hunting dogs. (<http://www.esccap.org>). Although close relationship between dogs and humans have their benefits, the presence in large number of stray dogs in Romania is problematic and may contribute to an enlarged reservoir for the disease. Public education is needed in order to inform and help prevent the spreading of zoonotic diseases, especially in countries

where efficient measures of control and prophylactic measures are deficitary.

The aim of the study was to establish epidemiological aspects of toxocarosis and associated risk factors in dogs from Cluj County, but also of other parasitic co-infections that can affect dogs from our region.

Material and methods

Study population

A total number of 155 fecal samples were analyzed from dogs aged between 2 weeks to 14 years during a period of 2 years in Cluj County. Animals were both pure breed and mixed breed.

All dogs presented at the veterinary clinics at the University of Agricultural Sciences and Veterinary Medicine from Cluj-Napoca for parasitological examination, either for a diagnosis or for a routine control. Owners were asked to fill in questionnaires regarding age, breed, sex, origin, deworming status in the last 3 months prior to the study, and absence or presence of clinical manifestations, such as vomiting, diarrhea, loss of appetite.

Parasitological procedures

The samples were taken in plastic recipients labeled accordingly and refrigerated until examination at the Parasitology and Parasitic Diseases Department. After macroscopic examination, fecal samples were analyzed using a flotation method with sodium chloride solution (sp.gr. 1.20). Approximately 5 grams of sample were mixed with the solution and examined after 30 minutes of flotation by light microscopy (x10, x20) for identification based on morphology. Species of *Isospora* genus were registered as *Isospora* spp.

Statistical analysis

Frequency, prevalence and its 95% confidence interval were calculated for total infection, each parasite, co-infection and studied variables. Chi-square test was used to establish the risk factors associated with canine toxocarosis. The studied variables were age, sex, origin, clinical

manifestations and deworming status. The age groups were represented by puppies (<3 months old), youth (between 4 and 12 months) and adults (over 1 year). According to origin, the dogs were divided in pets, kennel and foster. The differences among groups were considered significantly when *p* value was equal to or less than 0.05. The entire statistic was done with Epi Info™ 7 (CDC, USA) software (Dean et al., 2011).

Results

The overall prevalence of parasites was 67.1% (104/155; 95% CI: 59.3-74) with eight

different endoparasites identified (genus/species). *T. canis* (52/155; 34.8%; 95% CI: 25.9-41.5) was the most common parasite found in the examined samples. Other endoparasites found were *Trichocephalus vulpis* 20.0% (31/155), *Isospora* spp. 15.5% (24/155), *Ancylostoma caninum/Uncinaria stenocephala* 10.4% (16/155), *Giardia duodenalis* 7.1% (11/155) and less frequent (1/155) *Toxascaris leonina*, *Capillaria aerophila* and *Dipylidium caninum* (table 1).

Table 1. Prevalence and frequency of endoparasites parasites in 155 client-owned dogs

Endoparasites	Frequency	Prevalence %	95% CI
<i>T. canis</i>	54	34.8	25.9-41.5
<i>T. vulpis</i>	31	20.0	14.0-27.2
<i>Isospora</i> spp.	25	16.1	10.7-22.9
<i>A. caninum/U. stenocephala</i>	16	10.4	6.6-16.3
<i>G. duodenalis</i>	11	7.1	3.6-12.3
<i>T. leonina</i>	1	0.7	0.0-3.5
<i>C. aerophila</i>	1	0.7	0.0-3.5
<i>D. caninum</i>	1	0.7	0.0-3.5
Total	104	67.1	59.1-74.4

Table 2. Occurrence of single and mixed endoparasite infections in 155 client-owned dogs

Type of parasitic infection	No.	Prevalence %	95% CI
Single endoparasite infection	70	45.2	37.5-53.0
<i>T. canis</i>	31	20.0	14.5-27.0
<i>T. vulpis</i>	23	14.8	10.1-21.3
<i>G. duodenalis</i>	7	4.5	2.2-9.03
<i>Isospora</i> spp.	7	4.5	2.2-9.03
Other parasites	2	1.3	0.4-4.6
Mixed endoparasite infection	34	21.9	16.1-29.09
<i>T. canis</i> + <i>Isospora</i> spp.	13	8.4	5.0-13.8
<i>T. canis</i> + <i>T. vulpis</i>	3	1.9	0.7-5.5
<i>T. canis</i> + <i>Isospora</i> spp + <i>A. caninum/U. stenocephala</i>	3	1.9	0.7-5.5
<i>T. canis</i> + <i>A. caninum/U. stenocephala</i>	2	1.3	0.4-4.6
<i>T. canis</i> + <i>G. duodenalis</i>	1	0.6	0.1-3.6
<i>T. canis</i> + <i>D. caninum</i>	1	0.6	0.1-3.6
<i>T. vulpis</i> + <i>A. caninum/U. stenocephala</i>	4	2.6	1.0-6.5
Other co-infections	7	4.5	2.2-9.0

The prevalence for single and mixed parasitic infections was 45.2% (70/155) and 21.5% (34/155), respectively. *T. canis* was found in 20.0% (31/155) of cases as single infection and in 14.8% (23/155) of cases co-infection with other endoparasites. The most prevalent mixed endoparasite infection was between *T. canis* and *Isospora* spp. (8.4%, 13/155), but other five co-infections were observed (table 2).

Age was identified as a risk factor for *T. canis* infection ($p=0.000$). The highest prevalence (55.7%) was found in puppies (34/61; 95% CI:

42.5-68.5). Also, statistically significant difference ($p=0.000$) was obtained regarding the deworming status of the dogs. In dogs recently dewormed the prevalence was lower (12.3%) than in not dewormed dogs (51.1%). Neither origin, gender nor clinical manifestations were identified as risk factors (table 3). A slight increase in the rate of infection was seen in dogs from kennels (46.2%) and in foster dogs (36.8%), in comparison with pets (29.2%).

Table 3. Risk factors for *T. canis* in client-owned dogs from Cluj County

Variable		Frequency	Prevalence	95% CI	p
Age	Puppies (n=61)	34	55.7	42.5-68.5	0.000
	Youth (n=34)	12	35.3	19.7-53.5	
	Adults (n=60)	8	13.3	5.9-24.6	
Gender	Males (n=93)	31	33.3	23.9-43.9	0.8695
	Females (n=62)	23	37.1	25.2-50.3	
Origin	Pet (n=72)	21	29.2	19.0-41.1	0.3377
	Kennels (n=26)	12	46.2	26.6-66.6	
	Foster (n=57)	21	36.8	24.4-50.7	
Dewormed recently (<3 months)	Yes (n=65)	8	12.3	5.5-22.8	0.000
	No (n=90)	46	51.1	40.3-61.8	
Clinical signs	Yes (n=76)	29	38.2	27.2-50.0	0.5303
	No (n=79)	25	31.6	21.6-43.1	

Discussions

Numerous studies were conducted worldwide in order to evaluate the prevalence of toxocarosis in dogs. There are differences based on geographical location, with the values situated between 53.9% in Ethiopia (Jones et al., 2011) and 2% in the United States (Gates and Nolan, 2009). Few studies were carried out in Romania that focused on establishing the rate of infection of canine toxocarosis, but also the associated risk factors and co-infections. The results of our study revealed a high prevalence of dogs which shed *Toxocara* eggs (34.8%), being the most frequent parasite. A previous study from Romania reported an overall prevalence of 24.3% in 558 stray and owned dogs (Mircean et al., 2012). Other romanian authors observed an increased prevalence in shelter dogs (30%), rural owned dogs (47%), pets (17%) and owned dogs from private

clinics (17.5%) (Mureşan et al., 2002; Negrea, 2005; Amfim et al., 2011; Ardelean et al., 2005).

Similar results have been found in two central European countries such as Hungary and Poland (Poznan), where the prevalences were 30.1% (Fok et al., 2001) and 32.0% (Luty, 2001), respectively. In western and north Europe a much lower rate of infection is reported, 6.1% in Germany (Barutzki and Schaper, 2013) and 3.1% in Finland (Pullola et al., 2006).

The prevalence of *Toxocara canis* can be influenced by numerous factors, some of them identified as risk factors in other previous studies, as age, sex, breed, nutrition, geographical origin, habitat, coprophagy, owners' attitudes regarding deworming, dogs' immunity (Pullola et al., 2006; Mohamed et al., 2009; Fahrion et al., 2011; Nijse et al., 2015; Nijse et al., 2016). Age was determined as a

risk factor with half puppies positive to *T. canis* infection (55.7%). The lowest prevalence was obtained in dogs over one year old (13.3%). These results are in line with other studies (Szabová et al., 2007; Luty, 2001; Barutzki and Schaper, 2013). As in other studies (Nijse et al., 2015; Sahu et al., 2014), no differences were found between female and male dogs. Also, the origin of the dogs was not considered a risk factor ($p=0.3$), even a higher prevalence was identified in the kennels and in the fosters than in pets. In our study, these different values may be influenced by the age and deworming status in each group regarding the origin. The majority of kenneled dogs were aged under one year (73.1%, 19/26) and only 20.0% were registered as recently dewormed. In the case of fostered dogs, the number of animals was approximately equal for dogs under and over one year, but only 15% received anthelmintic treatment, as 64% of pets (60% young dogs and 40% adults) were dewormed regularly. A similar difference was noticed in the studies of Claerebout et al. (2009) in Belgium, which highlighted a prevalence of 26.3% in kenneled dogs, while only 4.4% of owned dogs were positive to *T. canis*. Fostered dogs had a high rate of infection also (34.6%). These results may suggest that the majority of the owners are more aware of preventing and treating parasitic diseases in companion animals, in contrast with the fostered ones that usually origin from shelters or streets.

Another risk factor was the deworming status of the dogs. From all 155 dogs, only 65 were dewormed in the last 3 months before presenting to the clinic. Highly significant differences in the rate of infection with *T. canis* were seen between dogs that recently received an anthelmintic therapy (12.3%) and those that were not dewormed in the last 3 months prior to the examination (51.1%) ($p=0.000$). In a study focused on risk factors of canine toxocarosis and deworming, Nijse et al. (2015), observed that only 24.0% of the owners followed a recommended therapeutic protocol and for 18.0% of them and that deworming dogs 3 or 4 times a year in agreement with ESCCAP, may reduce *Toxocara* eggs shedding.

We also focused on obtaining data about the health status of the dogs. Half of the dogs

presented clinical digestive signs with others being asymptomatic, but there were no differences between them regarding toxocarosis and shedding eggs (38.2% and 31.6%). These results emphasize the possibility of dogs being positive to *Toxocara canis* but not diagnosed and not receiving the proper treatment. Asymptomatic animals contribute to environment contamination by shedding eggs and subsequently to zoonotic risk. Therefore, performing regular coprological examinations and preventive deworming is advised, especially in dogs with higher risk of infection. Also, the zoonotic aspect of the disease, requires a more strict access of dogs in public places and especially a proper management of hygiene and cleaning-up the feces of dogs.

Most frequent mixed infection was between *T. canis* was *Isospora* spp. (8.4%). This can be due to the fact that age is a risk factor for both of the parasites., High prevalence of the co-infection was observed by other studies (Altreuther et al., 2011). We also found other co-infections with *A. caninum*/*U. stenocephala*, *T. vulpis*, *D. caninum* and *G. duodenalis*, some of them increasing the zoonotic potential of the parasitic infection with significant implication in public human health. These findings suggest the importance of diagnosis in parasitic diseases in dogs in order to give proper and efficient treatment for maintaining dogs' health.

Conclusions

Among the high rate of parasitic infection, toxocarosis had the highest prevalence in dogs in our study, especially in young animals and dogs that were not dewormed properly.

Recommendations

Canine toxocarosis represents a concern of both animal and public health and requires a proper management of efficient therapeutic and prophylactic measures. Further studies regarding the epidemiology of canine toxocarosis and the environmental parasitic pollution must be followed in our country to evaluate a more realistic prevalence and zoonotic potential of *T. canis*.

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