

Prevalence of intestinal parasitic infections in dogs from Havana, Cuba: risk of zoonotic infections to humans

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Abstract

Dogs harbour a variety of intestinal parasites, some of which have a negative impact in their health status and also have a relevant health-risk impact for human beings. The present study was conducted to determine the prevalence of intestinal parasitic infections in a population of stray and domestic dogs from Havana, Cuba. Serial stool samples from 293 dogs, derived in 178 stray dogs and 115 domestic dogs were analyzed by parasitological techniques, comprising direct wet mount, flotation and sedimentation techniques. The association between the infected dogs with the variables: age, sex and provenance (domestic or stray) were determined. In total, 168 dogs (57.3%) were infected at least with one intestinal parasite of veterinarian importance. The zoonotic helminths *Ancylostoma caninum* (45.4%) and *Toxocara canis* (25.3%) were the species more prevalent. *Cystoisospora* sp., (9.2%) was the protozoa infection more frequently identified. Infection rates were statistically significant in stray dogs than their counterparts, especially to infections caused by *A. caninum* and *T. canis*. There were not found significant differences regarding the sex of the canids with the infecting parasite. Puppies were more prone to infection with the majority of intestinal parasites identified compared with the adult dogs and had a high level of co infection as well. It is advisable to follow up the monitoring of faecal pollution with canine intestinal parasites in urban environments for understanding the dynamics of zoonotic helminths and make an integrated approach in order to minimize the risk of infection in different settings.

Introduction

Since antiquity dogs have played an important role as a source of companionship, emotional support and recreation to human around the world [1]. Despite this close association, human health can be compromised due to allergic reactions, trauma, and the transmission of more than 60 zoonotic infectious diseases some of which are due to canine intestinal parasitic infections [2].

Humans most commonly become infected with these zoonotic parasites through consumption of infected food or water or via direct fecal-oral contamination. The results of these infections may vary from asymptomatic carriage to long-term morbidity and even death. Among gastrointestinal parasites, dogs are hosts for several species, including wide spread parasites that affect humans such as *Ancylostoma caninum* (cutaneous larva migrans and eosinophilic enteritis) and *Toxocara canis*, which is a major health problem due to visceral migration and damage that may affect important organs, such as the eyes, liver and brain [3].

Intestinal parasites in canine population cause a clinical spectrum varying from subclinical to chronic state, which can deteriorate the animal health and the outcome in some cases can be fatal [4].

In Cuba there are estimates of two million of dogs, approximately one per six habitant, and half of this canine population are stray dogs according to data of the Ministry of Cuban Health, from which 200 000 resides in the capital of our country. Such free-ranging behaviour actually enhances parasite transmission between dogs, humans and wildlife.

Considering the lack of current knowledge about the prevalence of intestinal parasites in our country and aspects related to public and animal health, we aimed in this study to determine the prevalence of intestinal parasites among stray and domestic dogs in Havana, Cuba.

Material and method

Study area and analysed dogs

A descriptive cross-sectional study was carried out from October 2015 to March 2017 in a population of dogs from all municipalities of Havana province attending at the Canine Veterinary Unit. Using a prevalence of infections 10%, a total sample size of 293 out of 200.000 of total dogs was calculated for 90% confidence level.

A standard questionnaire was given to dog owners and veterinarians included in this study to obtain data on individual animals with respect to description, age, sex, breed, presence of ectoparasites and gregarious behaviour. Dogs were randomly selected (every three other dog) from those examined by a veterinarian with approximately half of the dogs drawn from the owned population and the other half from the stray population kennelled at the shelter. The final sample for this study comprised 178 stray dogs and 115 household dogs.

Coproparasitological analysis

Stool samples from domestic dogs were collected by their owners by rectal swab or collection of fresh emitted sample. Only shelter dogs kept in individual kennels were included to avoid potential cross-contamination during faecal collection. Faecal samples from both groups of dogs were collected in sterile containers and sent to the

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National Reference Laboratory of Intestinal Parasitic Infection from "Pedro Kouri" Institute for parasitological analysis.

Macroscopic examination was firstly performed for the detection of proglottids of cestodes or adults of *Toxocara*. Faecal samples were examined for intestinal parasites by a wet smear stained with Lugol's iodine and followed by the formalin ethyl acetate concentration technique. Samples also were examined using the Kato-Katz smear method and Willis-Malloy flotation technique [5]. In addition, all diarrheal faecal samples were stained by a modified acid-fast method for *Cryptosporidium* spp., and *Cystoisospora* [5]. The eggs, cysts, and oocysts found were identified according to morphological and staining characteristics under light microscopy.

Statistically analysis

All data were analysed using EPINFO 6.04 and EPIDAT 3.1 statistical programmes. For qualitative variables Chi square test and Fisher exact test were employed to assess the significance of the associations. The odds ratio (OR) with 95% confidence interval (CI) were performed as measures of association. The association between potential risk factors and intestinal parasitic infections was assessed by the Chi-square test with a 95% confidence interval. The P values less than 0.05 were considered as statistically significant for all test.

Ethical aspects

The research protocol (reference CEI-IPK-32-16) was approved by the Ethics Committee of the Pedro Kouri Institute. Each symptomatic

dog was seen and treated by a veterinarian professional involved in this study.

Results

From the 293 dogs examined, 168 turned out to be positive for intestinal parasites of medical importance for an overall prevalence of 57.3%. The zoonotic parasites *Ancylostoma caninum* (133 positive cases, 45.4%), and *Toxocara canis* (74; 25.3%) were the most prevalent. Other less frequently diagnosed parasites were *Cystoisospora* sp. (27; 9.2%), *Trichuris vulpis* (19; 6.5%), *Dipylidium caninum* (12; 4.1%), *Giardia lamblia* (10; 3.4%), *Eimeria* spp. (3; 1.0%) and *Blastocystis* spp. (2; 0.7%).

There were no significant differences in prevalence of intestinal parasitic infections observed between female and male dogs in this study, but with regard to sex, *Ancylostoma caninum*, *Toxocara canis* and *Cystoisospora* spp, were more frequently detected in young dogs, while *Trichuris vulpis* were more commonly diagnosed in dogs older than one year (Table 1).

From the 293 dogs investigated, 178 derived from stray dogs and 115 domestic dogs (Table 2). In total, 71.9% (CI: 65.0-78.8) of stray dogs were found harbouring at least one intestinal parasite of veterinarian importance and the odds ratio (OR) of infection was higher in shelter dogs than domestic dogs (OR=4.8; IC:2.9-7.9; P= 0,000+). The overall infection rate was 56.7% (CI: 49.2-64.3) for *Ancylostoma caninum* and 35.4% (CI: 28.1-42.7) for *Toxocara canis* as the most prevalent; whereas the overall infection rate in domestic dogs was 34.8% (CI: 25.6-43.9)

Table 1. Prevalence of intestinal parasites with regard to sex and age in dogs from Havana, Cuba

Intestinal Parasites	Prevalence, total %					
	Sex		P value	Age		P value
	Male (n = 142)	Female (n = 151)		< 1 year (n = 104)	>1 year (n = 189)	
<i>Ancylostoma caninum</i>	57 (40.1)	76 (50.3)	0.10	68 (65.4)	65 (34.4)	0.000+*
<i>Toxocara canis</i>	29 (20.4)	45 (29.8)	0.08	58 (55.8)	16 (8.5)	0.000+*
<i>Cystoisospora</i> sp.	12 (8.5)	15 (9.9)	0.69	17 (16.3)	10 (5.3)	0.004*
<i>Trichuris vulpis</i>	11 (7.7)	8 (5.3)	0.48	2 (1.9)	17 (9.0)	0.04*
<i>Dipylidium caninum</i>	7 (4.9)	5 (3.3)	0.56	6 (5.8)	6 (3.2)	0.44
<i>Giardia lamblia</i>	6 (4.2)	4 (2.6)	0.53	4 (3.8)	6 (3.2)	0.97
<i>Eimeria</i> spp.	2 (1.4)	1 (0.7)	0.61	1 (0.96)	1 (0.53)	0.76
<i>Blastocystis</i> spp.	1 (0.7)	1 (0.7)	0.96	1 (0.96)	1 (0.53)	0.76

*Statistically significant difference

Table 2. Prevalence of intestinal parasites in relation to the dog populations studied in Havana

Dog population	Parasite species	Prevalence (%) (95%CI)
Stray dogs (n=178)	All	71.9 (65.0-78.8)
	<i>Ancylostoma caninum</i>	56.7 (49.2-64.3)
	<i>Toxocara canis</i>	35.4 (28.1-42.7)
	<i>Cystoisospora</i> spp.	10.7 (5.9-15.5)
	<i>Trichuris vulpis</i>	6.7 (2.8-10.7)
	<i>Dipylidium caninum</i>	4.5 (1.2-7.8)
	<i>Giardia lamblia</i>	2.2 (0.6-5.7)
Domestic dogs (n=115)	All	34.8 (25.6-43.9)
	<i>Ancylostoma caninum</i>	27.8 (19.2-36.5)
	<i>Toxocara canis</i>	9.6 (3.8-15.4)
	<i>Cystoisospora</i> spp.	6.9 (1.9-12.0)
	<i>Trichuris vulpis</i>	6.1 (1.3-10.9)
	<i>Dipylidium caninum</i>	3.5 (0.9-8.7)
	<i>Giardia lamblia</i>	5.2 (0.7-9.7)

with these two zoonotic species as well, being the most commonly identified. The prevalence of infection by *Ancylostoma caninum* (OR=3.4; CI: 2.1-5.6) and *Toxocara canis* (OR=5.2; CI: 2.6-10.3) in stray dogs were statistically significant when comparing to domestic dogs.

With regard to age among the population studied, there was no significant difference between the rates of infection in domestic dogs when comparing the group of less than 1 year with the group above 1 year (Table 3). In the population of stray dogs, puppies had 12.3 more risk of infection than adults (OR=12.3, CI: 4.6-33.1; P= 0,000+). In summary, puppies from both groups, were more susceptible to intestinal parasitic infections (OR=6.2, CI: 3.5-11.2; P= 0,000+) than adult's one.

Co infection pattern were identified in 57/86 (66.3%) of infected dogs below 1 year, with *Ancylostoma caninum* and *Toxocara canis* mixed infection being the most common reported. In adult dogs the pattern of mono parasitism was the most prevalent in 61/82 (74.4%) dogs.

Discussion

Intestinal parasitic infections, particularly ascarids, hookworms, whipworms, *Giardia*, and *Cystoisospora* spp., are a common and important finding in dogs presenting to veterinarians [6]. Infections with intestinal parasites may result in some clinical symptoms and signs in dogs such as vomiting, diarrhoea, anaemia, anorexia, dermatitis, and loss of condition. In the present study we identified a prevalence of intestinal parasites of veterinarian importance in 57.3% of all dogs analysed, revealing a very high level of infection, similar to another study made in Argentina in which a prevalence of 52.4% was reported [7]. Other similar prevalence rates of intestinal parasites in dogs from Latin American countries show a 54% in Brazil [8], 35.5% in Venezuela [9], 52% in Argentina [10], and 64.8% in Chile [11].

The zoonotic helminths *Ancylostoma caninum* and *Toxocara canis*, were, by far, the most prevalent parasites identified. In addition, these species are responsible to its deleterious effects on dog's health for a wide variety of public health hazards. This result reveals that the risk of zoonotic diseases from dogs is strongly present in the areas where dogs came from, specially, if there is a lack of veterinary attention (Traub et al. 2005).

The most prevalent dog parasite identified in the present study *Ancylostoma caninum* is a widely distributed hookworm that locates in the small intestine of the dog and other canid species. The overall prevalence for *A. caninum* of 45.4% is in accordance with other studies [12,13,14]. It is known that females of *A. caninum* are very prolific and dogs can become infected in many ways (ingestion of free larvae from the environment, ingestion of larvae from paratenic hosts, maternal milk and transcutaneous penetration by infective larvae,) so it is presumed that it is almost impossible to prevent infection of dogs in endemic areas [6]. Particularly, in humans *A. caninum* can cause Cutaneous Larva Migrans (CLM). In Cuba CLM is mainly restricted to

case reports in dermatology consultation [15]. These patients typically present a history of prolonged exposure to wet surfaces in those public places where stray dogs are known to roam freely or antecedents of a recent visit to a beach. A relationship between the presence of *Ancylostoma* spp. larvae in soil of public squares and occurrence of cutaneous larva migrans in children has been demonstrated in Brazil [16].

Toxocara canis was the second intestinal parasite most frequently identified with a prevalence of 25.3%. Other recently studies made in Iran and in Poland have found comparable similar prevalence rates [17,18]. This nematode is very common intestinal helminths in domestic and wild canines, and it is also the agent of human toxocariosis [19]. In young pets this entity is recognized as an important public health hazard because it is responsible for the transmission of visceral and ocular larva migrans to humans in developing and developed areas around the world [20]. Recently, the problem of toxocariosis in humans and dogs has been highlighted worldwide [17]. In Cuba toxocariosis has frequency reported in children, especially with allergic diseases. In a previous study of 958 Cuban school-aged children a seroprevalence of 40.1% to *Toxocara* by ELISA screening was determined [21]. In fact, children are the subjects at highest risk of infection, due to exposure to areas (e.g. sandpits, green areas, gardens, playgrounds) potentially contaminated by *T. canis* eggs [22].

The coccidian *Cystoisospora* spp., was the most frequently identified among intestinal protozoan with a prevalence rate of 9.2%. This parasite is mostly found in puppies, and causes gastrointestinal signs in this population, as diarrhoea and loss of weight [23]. A similar prevalence rate was reported in Italy and Spain [23,24].

The percentage of dog samples in which whipworm eggs were detected in the present study was 6.5%. *T. vulpis* is not included in zoonotic intestinal nematodes of pets and its zoonotic potential is questioned although presumed cases of visceral larva migrans and of patent intestinal infections have been described in people (Traversa, 2011). Similar rates of whipworm infection in dogs have been published by other authors [18,25].

A significantly high proportion of young dogs were infected with *Ancylostoma caninum*, *Toxocara canis* and *Cystoisospora* sp. Some age-dependent analyses have shown that puppies are more susceptible to these intestinal parasites than adult dogs [6,26]. Particularly, younger dogs are more exposed to *Toxocara canis* and *Ancylostoma caninum* infections because they can acquire these helminths by several routes such as transmammmary routes by migrating larvae, ingestion of embryonated eggs from the environment or finally by somatic larvae via paratenic hosts [6]. Besides, in the epidemiology of toxocariosis in dogs, puppies are usually born with or acquire this ascarid infection early in life through trans-mammmary and trans-placental transmission [26].

As expected, infections caused by *Trichuris vulpis* were more frequently detected in adult dogs. The absence of a vertical transmission in *T. vulpis*, its long pre-patent period and a partial ability to stimulate a protective immune response, explain the more likely to be infected with this intestinal parasite in adult dogs rather than in puppies [10].

Although sex was not a significant risk factor in this study, female's dogs were more infected with *A. caninum* and *T. canis*, the primary species of roundworms and hookworms infecting dogs worldwide, than their male counterparts. This result may be due to the physiological

Table 3. Overall prevalence of parasitic infections in relation to the age group in the canine population studied

Kind of dogs	Puppies (< 1 year)		Adults (> 1 year)		P value
	Total	Infected (%)	Total	Infected No. (%)	
Domestic dogs	25	12 (48.0)	90	28 (31.1)	P= 0.18
Stray dogs	79	74 (93.7)	99	54 (54.5)	P= 0.000+
Total	104	86(82.3)	189	82(43.4)	P= 0.000+

peculiarities of the female dogs, which usually constitute stress factors thus reducing their immunity to infections [27]. Another point is that female dogs are relevant source of infection for other animals and environmental contamination because they often harbour somatic larvae, which mobilize during pregnancies and infect subsequent litters even when re-infections do not occur [2].

Stray dogs had a higher prevalence of all intestinal parasites compared to domestic dogs, with the exception of *Giardia lamblia*. It is known that infection rates of intestinal parasites in stray dogs are very high, especially to hookworm and ascarid eggs, because of their free-roaming behaviour, lack of veterinarian assistance and hence, no anthelmintic treatment, environmental contamination with parasite eggs has likely already occurred over a fairly dispersed area, resulting in the presence of infectious stages that pose a risk of infection to stray dogs [28].

Parasitic burdens and egg outputs are higher in puppies, but patent intestinal infections may occur in dogs of all ages and categories [2,29]. In this study we found that puppies of stray dogs had a higher rate of infection compared with adult stray dogs. In the same way, puppies of domestic dogs had a major prevalence of intestinal parasites when comparing with the adult counterparts. Puppies may become infected in utero and via the milk, but a proportion of mobilized larvae reach adulthood in the intestine and cause a patent infection with a long-lasting high egg shedding [2].

It is necessary that veterinary care and public health education need to be increased in order to protect the dogs, and also their owners. It should emphasize about the "One Health" concept by linking human and animal health [2] and increase the efforts in appropriate control programs towards a reduction of intestinal parasites in dogs in order to prevent the diffusion of zoonotic parasites in public areas. Owner should be educated to collect dropping voided by their own pets on public areas and to check the parasitic status of their dogs regularly. In addition, the parasitic monitoring of dogs younger than 12 months is a strong necessity for pets' owners.

In conclusion, the results obtained in this study suggest that intestinal parasitic infections are prevalent; specifically, those caused by *Ancylostoma caninum* and *Toxocara canis*. Stray dogs were more prone to be infected by helminths and protozoan parasites than domestic dogs. Puppies were at a higher risk of *A. caninum* and *T. canis* infections. Concerted efforts should therefore be made to embrace modern dog disease control programs and specifically the need for routine deworming of their dogs along with the stray dogs.

Competing interests

The authors declare that they have no competing interests.

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